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Synthesis of Second-Generation Sansalvamide A Derivatives: Novel Templates as Potential Antitumor Agents

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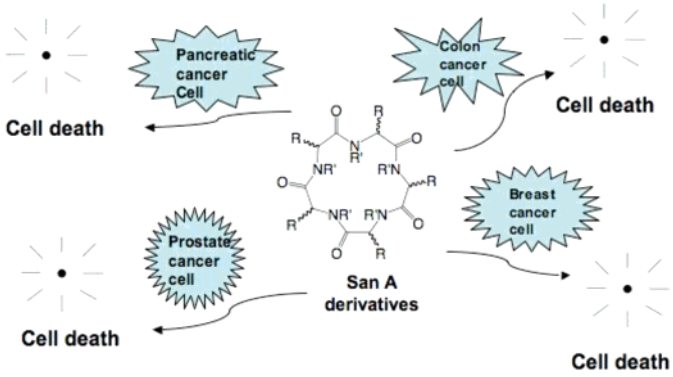
Received September 4, 2006



We report the synthesis of 34 second-generation Sansalvamide A derivatives. San A derivatives have unique anticancer properties and target multiple cancers, including colon, pancreatic, breast, prostate, and melanoma. As novel templates, the derivatives described herein explore the role of stereochemistry, amide bond geometry, transannular hydrogen bonding, and polarity on antitumor potency. Testing the chemotherapeutic activity of these derivatives against multiple cancer cell lines will provide clear structural motifs and identify conformational space that is important for cytotoxicity. The 34 compounds presented are divided into six series, where five series involve the insertion of D-amino acids in conjunction with four structural features at each of the five positions of the macrocycle. The sixth series involves comparison between all L- and all D-amino acid derivatives with *N*-methyls placed at each position around the macrocyclic core. The four structural features explored in conjunction with D-amino acids include *N*-methyl amino acids, aromatic amino acids, polar amino acids, and hydrophobic alkyl amino acids.

Introduction

Natural products are excellent sources of potential new drug leads. These novel structures are important for the development of original therapeutic leads that target new biological pathways. Sansalvamide A (San A) is one such natural product (Figure 1). San A, which is a depsipeptide isolated from a marine fungus (*Fusarium* spp.), exhibits antitumor activity.[1−3] A limited number of analogues have been made; to date, ∼46 analogues have been reported, where 36 were reported by our laboratory.[4,5] These San A derivatives have demonstrated that they are privileged structures and exhibit potency against multiple targets in numerous cancer cell lines. Examples of potent cytotoxicity against pancreatic,[4,6] colon,[3,5,7,8] breast, prostate, and melanoma

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10.1021/jo061830j CCC: $37.00
Published on Web 02/22/2007

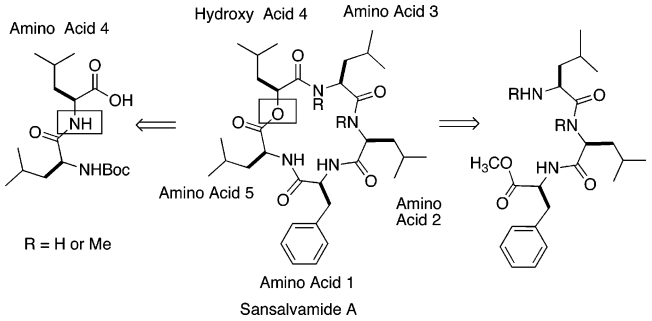
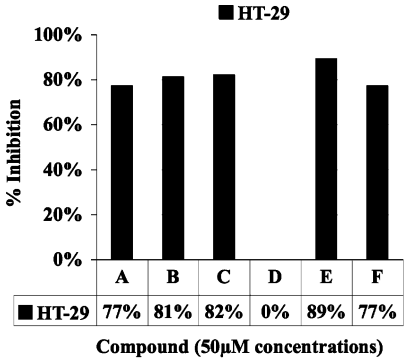


FIGURE 1. Retrosynthetic strategy.

cancers[4] clearly indicate the potential of this compound class as a platform useful in targeting these cancers.

Syntheses and evaluation of all-peptide analogues against colon cancer cell line HCT-116, a drug-resistant cancer cell line, have revealed two potent derivatives.[4,8,9] Previous work by our group has shown that five first-generation derivatives are potent against the drug-sensitive colon cancer cell line HT-29,[5,7] and two demonstrate potency against two drug-resistant colon cancer cell lines. Further, two first-generation compounds were found to be potent against three pancreatic cancer cell lines.[6] It was the structures of these six active compounds found in the first generation that were used as the basis for the design of second-generation compounds (Figure 2). Assaying a diverse set of San A analogues against multiple cancer cell lines will provide new structures as potential antitumor agents against individual cancers. This is particularly important given the lack of chemotherapeutic options for patients with pancreatic and drug-resistant colon cancers.[10−14]

Herein we describe the synthesis of 34 second-generation San A derivatives. The derivatives have been expressly designed from the first generation's cytotoxicity data and will provide valuable information on how stereochemistry, amide bond geometry, and polarity impact cytotoxicity. These second-generation derivatives also have a lower C log P average (2.2) than the first generation (∼2.9).[15] Thus, they should show improved ability to enter cells over the first-generation ana-



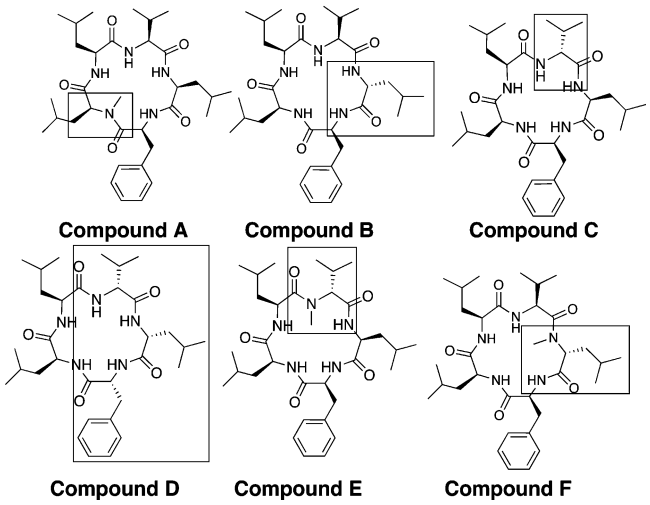


FIGURE 2. First-generation derivatives found to be cytotoxic against drug-sensitive cancer cell lines. Note, compound **D** was found to be cytotoxic against pancreatic cancers.

logues. Testing the chemotherapeutic activity of these derivatives against multiple cancer cell lines will provide clear structural motifs and identify conformational space that is important for cytotoxicity in multiple cancers. The recent success of San A derivatives targeting numerous cancers emphasizes the importance of the derivatives described within as potential new lead structures.

Results

Synthetic Strategy. San A is composed of four L-amino acids and one hydroxy acid. We report here the synthesis of 34 San A peptide analogues, involving the exchange of the hydroxy acid in position 4 to an amino acid (Figure 1). We used a succinct solution-phase synthesis route because of the hydrophobic nature of the residues. Our convergent approach, involving two fragments (Figure 1),[8] is amenable to inserting L- and D-amino acids systematically within San A. This route was also designed to facilitate large-scale synthesis for extensive biological studies.

Synthesis. Synthesis of 34 San A derivatives was completed using amino acids shown in Figure 3 via the synthetic route outlined in Scheme 1. Using 2-(1-*H*-benzotriazol-1-yl)-1,1,3-tetramethyluronium tetrafluoroborate (TBTU) and diisopropylethylamine (DIPEA), acid-protected residues **1a**−**d** and N-Boc-protected residues **2a**−**g** (Scheme 1) were coupled to give the dipeptides **1**−**2**-Boc (90−95% yield). Deprotection of the amine on residue **2** using TFA gave the free amines **1**−**2** (∼quantitative

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(15) The *C* log *P* values were calculated using an algorithm. The log *P* value of a compound, which is the logarithm of its partition coefficient between *n*-octanol and water log($c_{octanol}/c_{water}$), is a well established measure of the compound's hydrophilicity. Low hydrophilicities and therefore high log *P* values cause poor absorption or permeation. It has been shown for compounds to have a reasonable probability of being well absorb their log *P* value must not be greater than 5.0. The distribution of calculated log *P* values of more than 3000 drugs on the market underlines this fact.

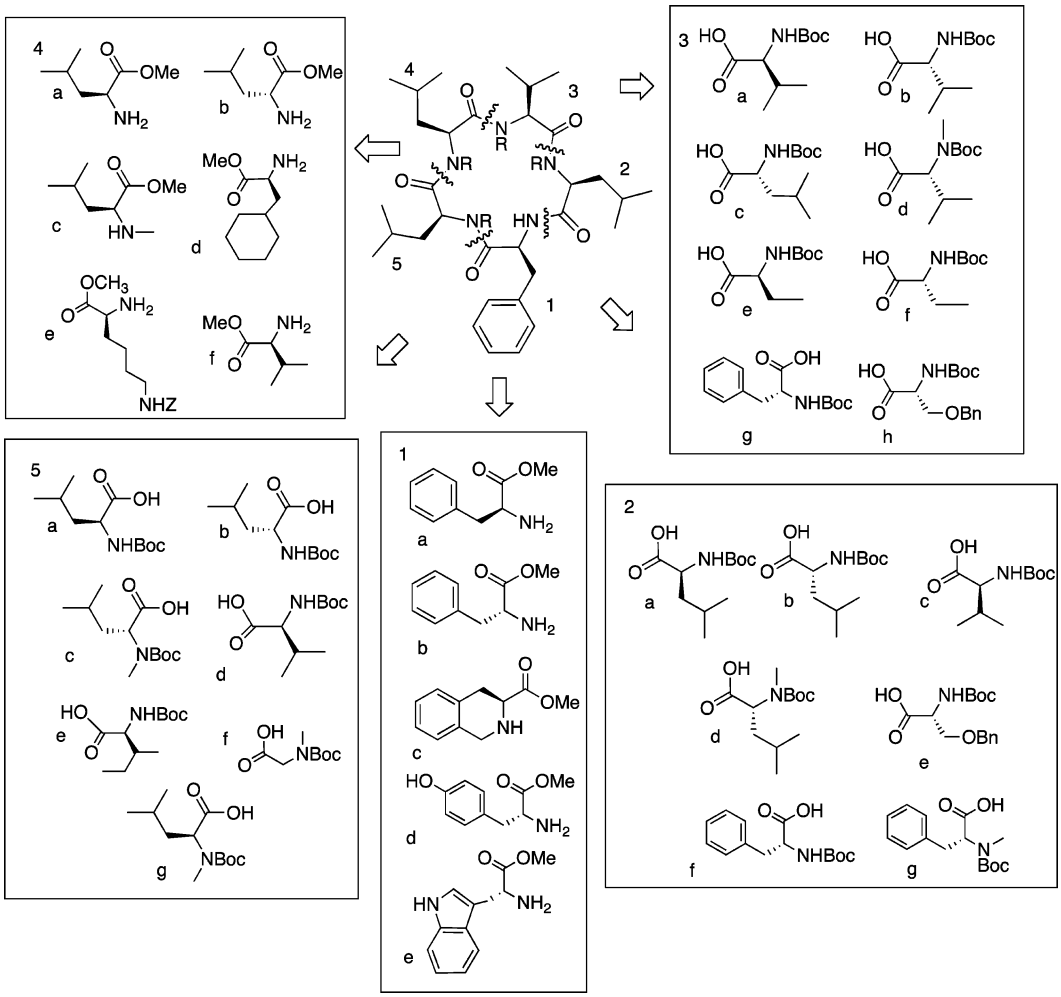

FIGURE 3. Amino acids used in San A derivatives.

yields). Coupling of this dipeptide to monomers **3a**−**h** gave the desired tripeptides (Fragment 1) in good yields (80−95%).[16] The synthesis of Fragment 2 was completed by coupling residues **4a**−**f** to residues **5a**−**g** to give the dipeptides **4**−**5**-Boc (90−95% yield). The amine was deprotected on Fragment 1 using TFA, and the acid was deprotected in Fragment 2 using lithium hydroxide. Fragments 1 and 2 were coupled using multiple coupling agents[8,17−19] yielding 34 examples of linear pentapeptides (70−90% yield).[16]

Cyclizing large macrocycles is usually very challenging, and typically the yields are low. The recent discovery of high-yielding conditions[20] provided the majority of macrocycles in good yields. Dissolving the linear pentapeptide in THF (0.05 M), addition of 2 equiv of anisole, and approximately 8 drops of concentrated HCl per 0.3 mmol of linear pentapeptide led to partially deprotected amine within 24 h. Four drops of concentrated HCl per 0.3 mmol of peptide was added. The reaction was allowed to stir at room temperature and was then checked after 24 h by LCMS. Typically deprotection of the acid and amine was complete within 4 days.[21] Upon completion, the reaction was concentrated in vacuo and dried on the high-vac. The dried, crude, free amine/free acid linear pentapeptide was dissolved in a 2:2:1 ratio of THF/CH_3CN/CH_2Cl_2 (0.004 M).[22] DIPEA (6 equiv) and three coupling agents [2-(1*H*-7-azabenzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate (HATU), 3-(diethoxyphosphoryloxy)-3*H*-benzo[*d*][1,2,3]triazin-4-one (DEPBT), and TBTU (0.7 equiv each)] were added to the reaction to yield a clear solution. Reactions were usually complete within 4−6 h.[23] The final products were generated after a standard workup utilizing ammonium chloride, extraction with methylene chloride, concentration in vacuo, purification via flash chromatography, and subsequent HPLC purification (yields ranged from 30 to 90% depending on the substrate).[24]

(16) Dipeptide and tripeptide structures were confirmed using ^{1}H NMR. All linear pentapeptides were confirmed using LCMS and ^{1}H NMR. (Note: ^{1}H NMR were taken for cyclized peptides, but due to their complexity, they were not seen as the primary confirmation for cyclized compounds.) See Supporting Information for spectra.

(17) Unpublished results from the Guy lab at Department of Chemical Biology and Therapeutics, St. Jude Children's Research Hospital, Memphis, TN 38103, and published results from our lab show that the use of several coupling reagents facilitates formation of the peptide bond in high yields.

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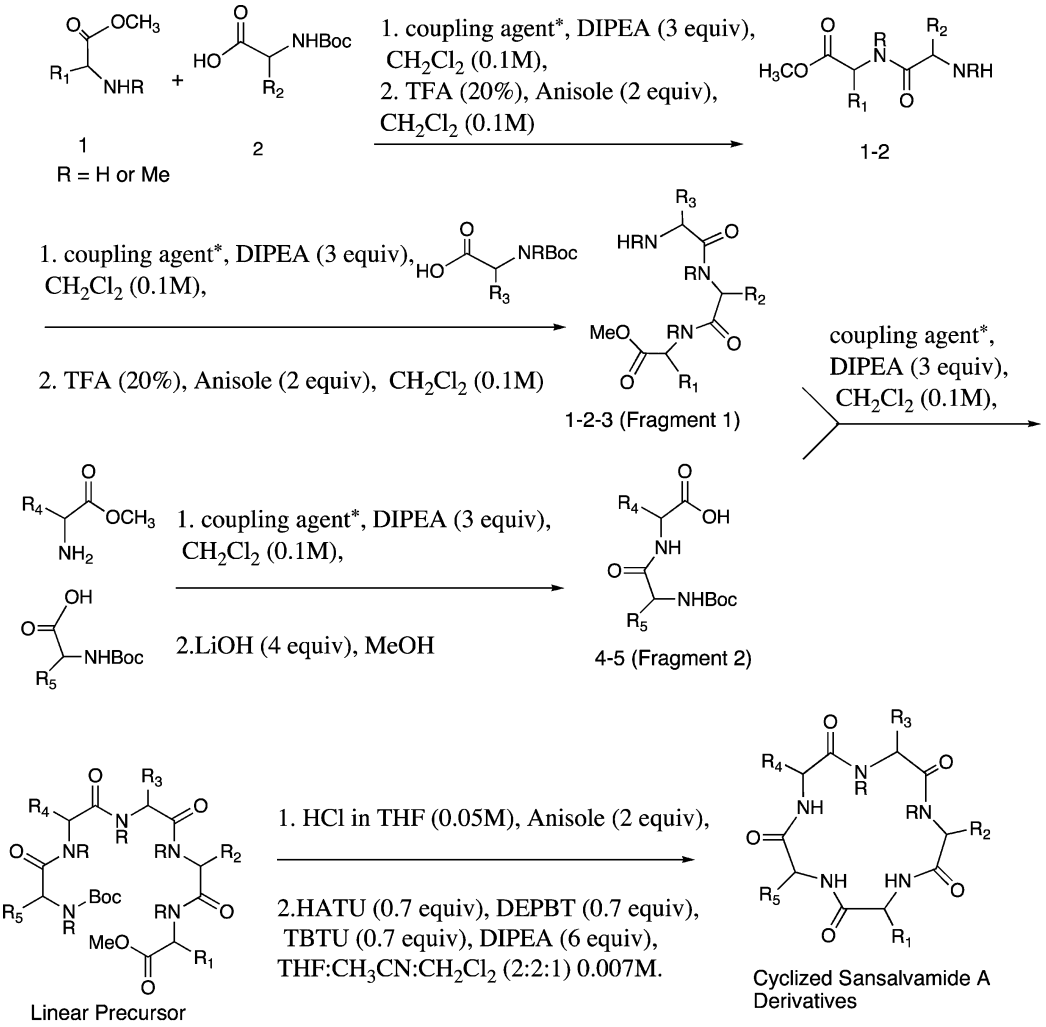
(21) For details on the reaction conditions see Supporting Information.

(22) The dodecapeptide tends to form at cyclization concentrations greater than 0.005 M. We found that cyclizing the linear pentapeptide at 0.004 M led to almost exclusively the pentapeptide macrocycle as the product.

(23) It was straightforward to follow the reactions via LCMS as the starting material double deprotected linear precursor would appear at 5.0−5.5 min and the cyclized product would appear between 6.1−7.0 min.

SCHEME 1. Synthesis of San A Macrocyclic Derivatives



* TBTU (1.2equiv), and/or HATU (0.75 equiv)[19]

Structures of Macrocyclic Derivatives. The second generation of San A derivatives was divided into six series. These series explore the impact of stereochemistry using D-amino acids in conjunction with four structural features: *N*-methyl amino acids, aromatic amino acids, polar amino acids, and hydrophobic alkyl amino acids. They include derivatives with alterations at position 1 (Figure 4), position 2 (Figure 5), position 3 (Figure 6), position 4 (Figure 7), position 5 (Figure 8), and comparison of all D- and all L-amino acids in conjunction with *N*-methyl moieties (Figure 9).

Figure 4 describes the alterations at position 1, where the first-generation compounds are shown in gray and the second-generation compounds reported here are shown in black. Compound **1** was designed as a comparison to San A peptide to determine if rigidifying the core structure using a conformationally restricted hydrophobic aromatic moiety would improve cytotoxicity. In the first generation, only compound **G** demonstrated any antiproliferation activity. Compounds **2** and **3** were both designed to follow up on the cytotoxicity observed with compound **G**, where aromatic D-amino acids with hydrogen-bonding elements were utilized. As with compounds **2** and **3**, compounds **4** and **5** were designed to explore the importance of a D-amino acid in position 1, but they also involved placement of a D-amino acid in several other positions shown to be important for cytotoxicity. Compound **4** contains a D-amino acid in positions 1 and 5 as well as an *N*-methyl moiety in position 4. Compound **5** contains D-amino acids in positions 1, 3, and 4. The primary modifications to these structures involved three of four structural features; in addition to inserting D-amino acids, we included an *N*-methyl moiety **4**, insertion of the tetrahydroisoquinoline as an alternative aromatic amino acid (compound **1**), and addition of polar aromatic D-amino acids (compounds **2** and **3**).

Figure 5 describes compounds with changes at position 2, where the first-generation compounds are shown in gray and the second-generation compounds reported here are shown in black. In the first generation, compounds **B** and **F** demonstrated the greatest antiproliferation activity of this series in position 2 (Figure 2).[5] All seven second-generation compounds contained D-amino acids in position 2 because the two active compounds in the first generation contained D-amino acids in this position. The difference between the two active first-generation compounds is an *N*-methyl moiety; therefore, we designed this next

(24) The 34 macrocyclic peptides have LCMS spectra given in the Supporting Information. In addition, 29 compounds out of 34 in the library have each intermediate characterized via NMR and/or LCMS.

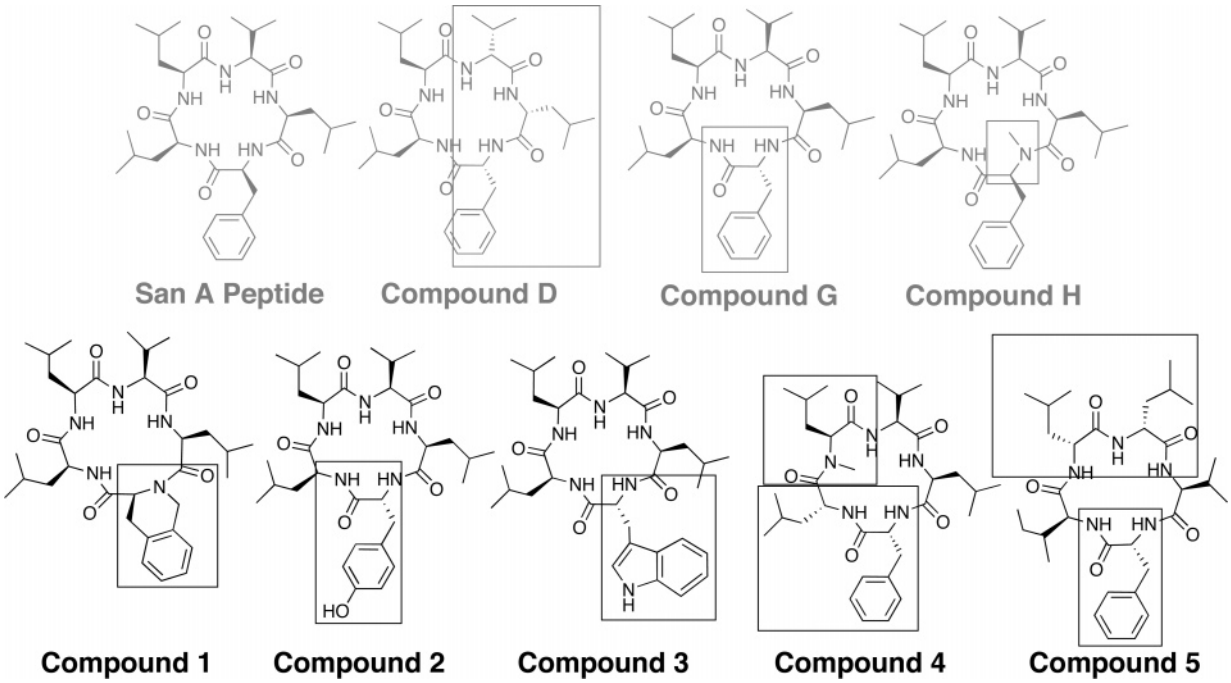


FIGURE 4. Compounds with changes in position 1: gray compounds are first generation (San A peptide, **D**, **G**, **H**); and black compounds are second-generation compounds (**1**, **2**, **3**, **4**, and **5**).

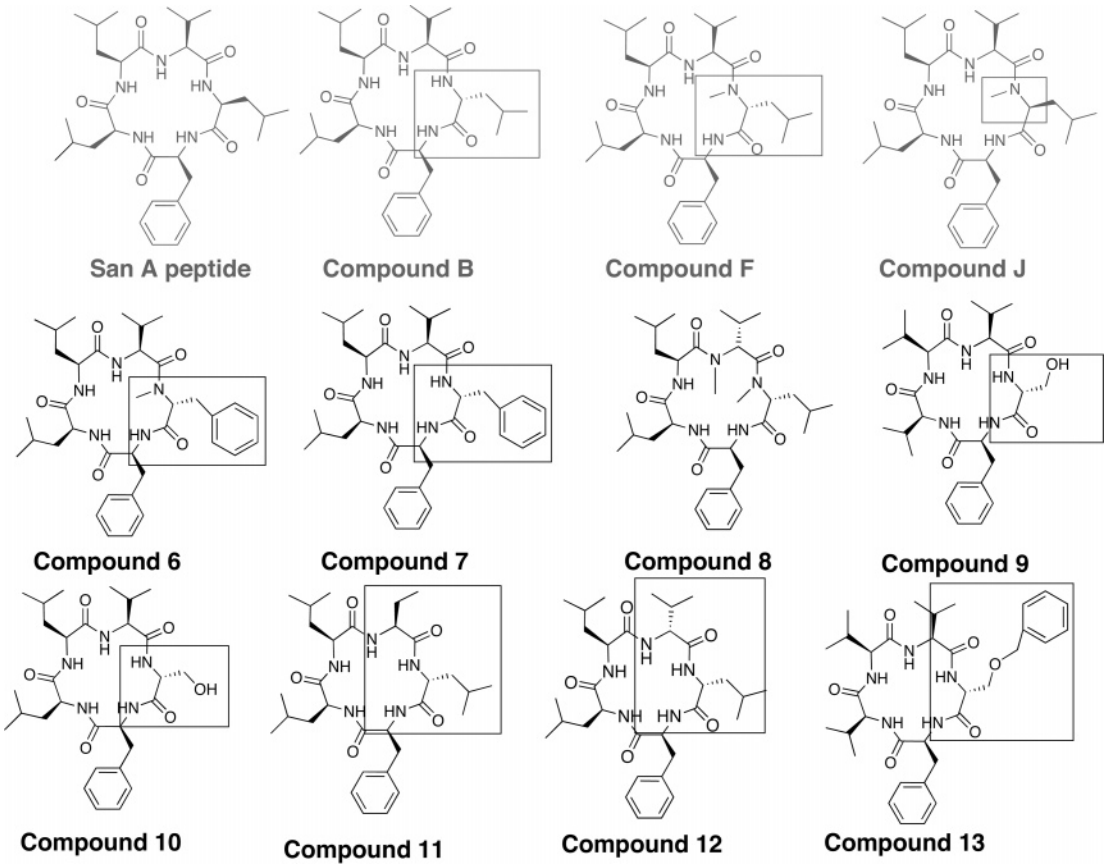


FIGURE 5. Compounds with changes in position 2: gray compounds are first generation (San A peptide, **B**, **F**, and **J**); and black compounds are second-generation compounds (**6**, **7**, **8**, **9**, **10**, **11**, **12**, and **13**).

generation to contain a D-amino acid both with (compound **6**) and without (compound **7**) an *N*-methyl moiety. In addition, given that two of the five most potent compounds in the first generation were compounds **E**, which contain an *N*-methyl D-amino acid at position 3, and **F**, which has an *N*-methyl D-amino acid in position 2 (Figure 2), we designed compound

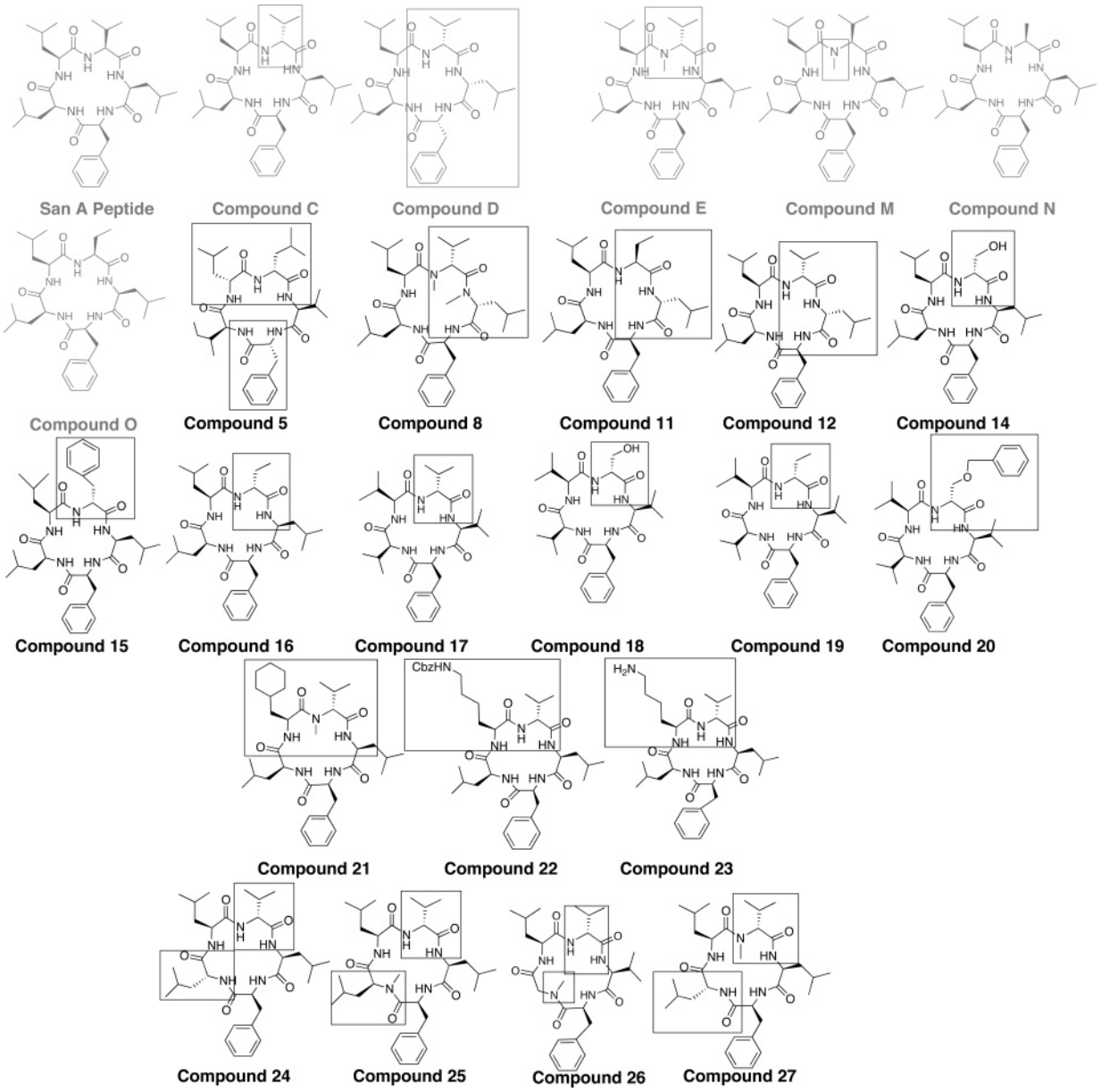


FIGURE 6. Compounds with changes in position 3: gray compounds are first generation (San A peptide, **C**, **D**, **E**, **M**, **N**, and **O**); and black compounds are second-generation compounds (**5**, **8**, **11**, **12**, **14**, **15**, **16**, **17**, **18**, **19**, **20**, **21**, **22**, **23**, **24**, **25**, **26**, and **27**).

8. Compound **8** contains an *N*-methyl D-amino acid at positions 2 and 3, and therefore, the results from cytotoxicity assays would indicate whether the insertions of these two amino acids had a synergistic effect. Further, compounds **B** and **C** were also two of the most biologically active compounds, and they contained a D-amino acid in positions 2 and 3, respectively; therefore, we designed compound **12**. Compound **12** explores the synergistic effect of placing D-amino acids in both positions but without the *N*-methyl moieties present.

We also synthesized two derivatives containing hydroxyl moieties at position 2 (compounds **9** and **10**) in order to examine whether a polar amino acid containing a H-bonding element at this position would improve its cytotoxic effect. In addition, we had noticed a trend that the conversion of a leucine to a valine residue sometimes promoted cytotoxicity, perhaps due to a lower overall molecular weight. Thus, we explored this hypothesis by synthesizing derivative **10**, which contains a serine in position 2 and leucines in positions 4 and 5 (which parallels those in the San A peptide) as well as **9**, which contains a serine in position 2 and valines in positions 4 and 5. Compound **11** was designed with the D-leucine seen in the active first-generation compound **B** at position 2, as well as an ethyl side chain replacing an isopropyl in position 3. This was designed to explore the structural requirements for potency at both positions 2 and 3. Finally, compound **13** involved placement of a D-benzyl-protected serine at position 2. This derivative will very effectively explore the impact of a hydrophobic aromatic residue at this position. Thus, the primary modifications of these structures involved three of the four structural features: inclusion of D-amino acids with *N*-methyl moieties, the addition of aromatic amino acids, and the insertion of polar residues into the San A backbone at position 2.

Figure 6 describes compounds with changes at position 3, where three of these first-generation compounds (gray: **C**, **D**,

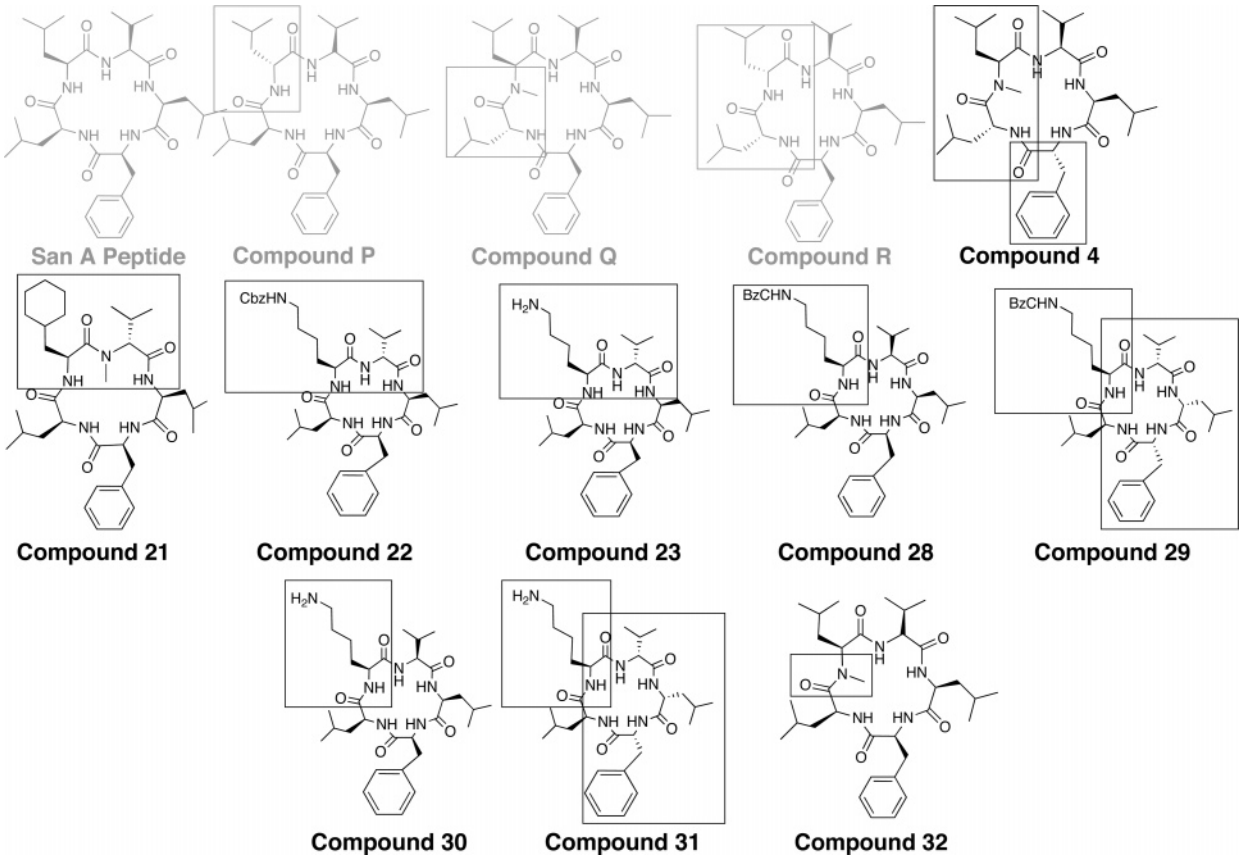


FIGURE 7. Compounds with changes in position 4: gray compounds are first generation (San A peptide, **P**, **Q**, and **R**); and black compounds are second-generation compounds (**4**, **21**, **22**, **23**, **28**, **29**, **30**, **31**, and **32**).

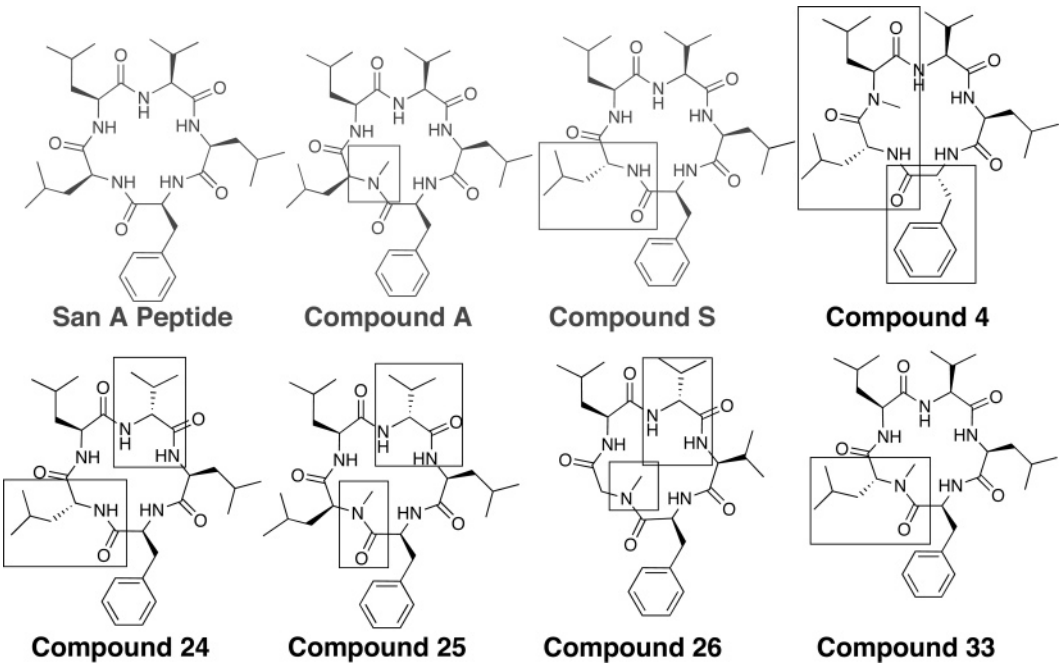


FIGURE 8. Compounds with changes in position 5: gray compounds are first generation (San A peptide, **A**, and **S**); and black compounds are second-generation compounds (**4**, **24**, **25**, **26**, and **33**).

and **E**) were the most cytotoxic of the first-generation San A series. Thus, we focused the design of our second-generation compounds on making changes to this position. In the first generation, compounds **C** and **E** demonstrated the greatest antiproliferation activity against colon cancer cell lines.[5] Given that these two active compounds contained D-amino acids in position 3 and were identical except that **E** contained an *N*-methyl moiety combined with a D-amino acid at this position,

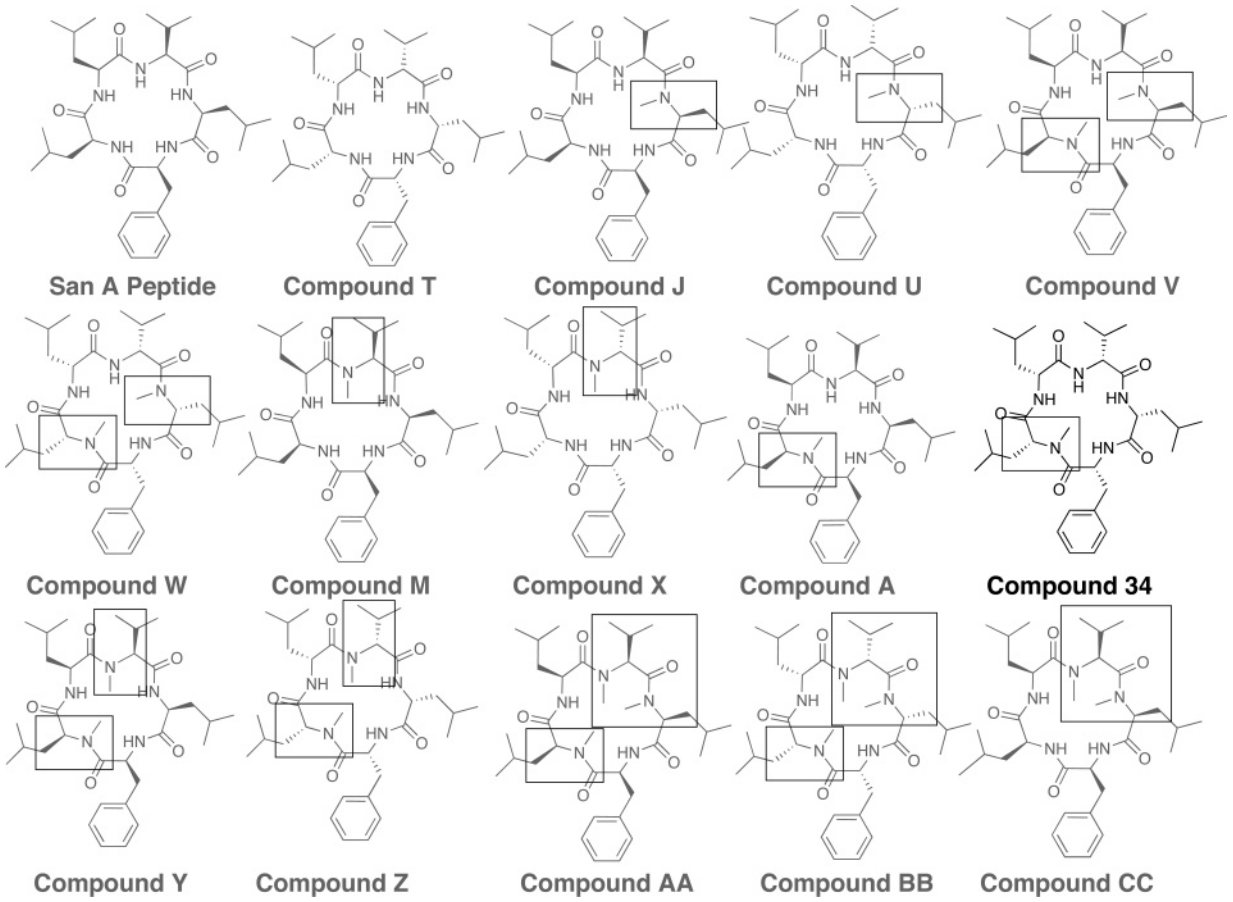


FIGURE 9. Compounds with combinations of D-amino acids and *N*-methyl moieties: gray compounds are first generation (San A peptide, **T**, **J**, **U**, **V**, **W**, **M**, **X**, **A**, **Y**, **Z**, **AA**, **BB**, and **CC**); and the only second-generation compound is shown in black (**34**).

we focused our attention on exploring this structural feature. Thus, 16 of the 17 second-generation compounds that explored the impact of position 3 contained D-amino acids in this position.

The design rationale for compounds **5**, **8**, **11**, and **12** was explained previously. The reason behind the synthesis of compounds **14**, **15**, and **16** was to explore the impacts of a polar moiety (serine), an aromatic moiety, and an ethyl moiety being placed in position 3, respectively. Compounds **17**, **18**, **19**, and **20** involve the conversion of L-leucines to L-valines in positions 2, 4, and 5 in order to reduce the molecular weight of the molecule while maintaining the steric bulkiness of the side chain. In addition to these changes, we also explored substitutions of D-amino acids at position 3, where compounds **17** and **19** both involved the substitution of an alkyl chain (isopropyl and ethyl, respectively). Compounds **18** and **20** involved the substitution of a polar moiety (serine) and an aromatic moiety (i.e., a benzyl-protected serine). Three compounds (**21**, **22**, and **23**) in this series explored alterations to positions 3 and 4. Compound **21** involved the substitution of an *N*-methyl D-amino acid in position 3 and a cyclohexyl moiety in position 4, which provides a hydrophobic element in position 4. Compound **22** contains a D-amino acid in position 3 and a carboxybenzyl-protected lysine in position 4, which incorporates an additional aromatic element into the San A backbone. Compound **23** has a D-amino acid in position 3 and a lysine in position 4, thus inserting a polar element into the backbone.

In addition to these three compounds that involve modifications at both the 3 and 4 positions, modifications to the 3 and the 5 positions were made in four San A derivatives (**24**, **25**, **26**, and **27**). Because a first-generation derivative with a D-amino acid in either the 3 or 5 position (**C** and **S**, respectively) was potent in the cytotoxicity assays, a derivative containing both a D-amino acid in positions 3 and 5 was synthesized, **24**. An active first-generation derivative incorporated an *N*-methyl at position 5 (**A**); therefore, we included a D-amino acid at position 3 into **25** and **26**. Compound **25** contains an *N*-methyl L-leucine at position 5, while compound **26** contains an *N*-methyl glycine. Finally, in order to determine whether there is a synergistic effect, compound **27** contains two features found to be independently important in cytotoxicity: an *N*-methyl D-amino acid at position 3 and a D-amino acid at position 5. In summary, our structural modifications at position 3 involved all four features, where the inclusion of *N*-methyl moieties, additional aromatic amino acids, polar moieties, and hydrophobic moieties provide compounds that allow us to optimize the structural features required for potency.

On the basis of the cytotoxicity assays of the first-generation structures, changes at position 4 appear to have a limited impact on activity; therefore, the only compounds synthesized in this series were made to determine whether modifications that would improve solubility properties or allow attachment of a biotin tag could be tolerated. Shown in Figure 7 are the nine compounds generated in this series. The design rationale for compounds **4**, **21**, **22**, and **23** was explained previously. Similar to **22**, compounds **28** and **29** both contained a carboxybenzyl (CBz)-protected lysine. While **22** contains a D-amino acid at

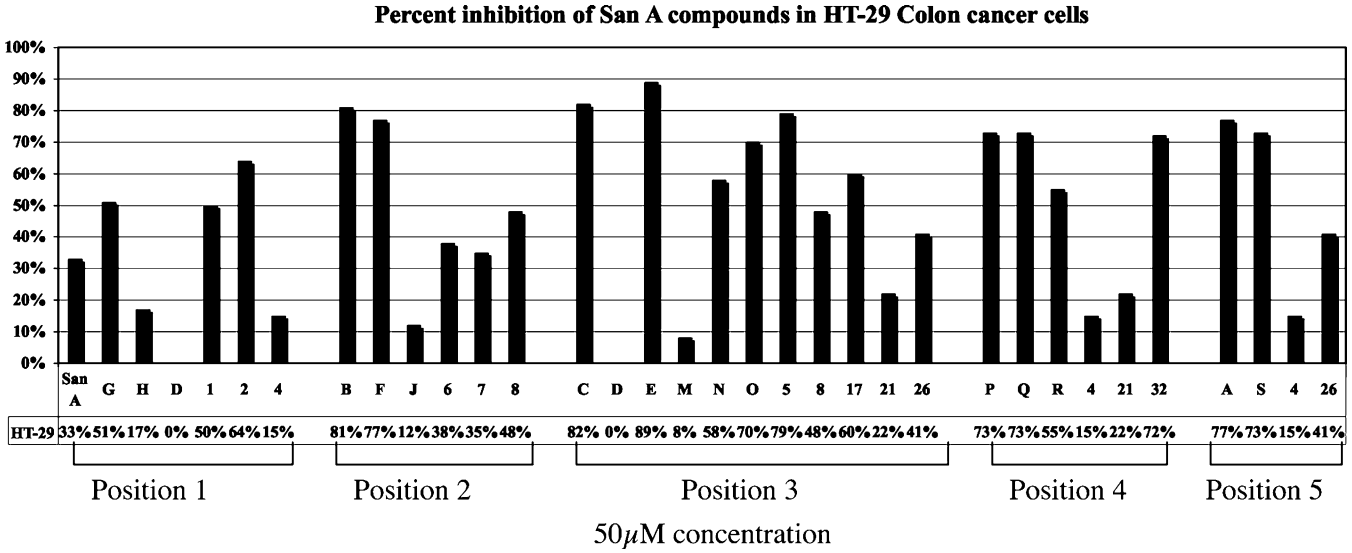


FIGURE 10. First- and second-generation compounds tested in colon cancer cell line HT-29 using thymidine uptake assays. All compounds were tested in three separate assays, in triplicate each time (i.e., nine data points were taken for each percent inhibition given). Error = ±2%.

position 3, compound **28** does not, rather it contains all L-amino acids. Thus **28** is identical to compound San A peptide with the exception of the CBz-protected lysine. Compound **29** was based on the cytotoxic compound **D** and, therefore, possesses three D-amino acids in positions 1, 2, and 3 in addition to the CBz-protected lysine at position 4. Removal of the CBz group from compounds **28** and **29** affords compounds **30** and **31**, respectively, thus providing the free lysine at position 4. These two compounds not only allow us to determine the impact of a polar moiety at this position, but they offer a handle upon which we can couple a biotin tag for affinity purification assays. Finally, compound **32** possesses an *N*-methyl moiety at position 4. Thus, our structural modifications at position 4 involved all four features, where the inclusion of *N*-methyl moieties, additional aromatic amino acids, polar moieties, and hydrophobic moieties will provide compounds that will allow us to explore the structural features required for potency.

On the basis of the cytotoxicity assays of the first-generation structures, an *N*-methyl at position 5 appears to improve potency, where compound **A** was one of the six most active compounds in the first generation (Figure 2). Therefore, three of the five second-generation compounds contain an *N*-methyl moiety at position 5 (Figure 8). In addition, compound **33**, which contains a D-amino acid at position **5**, demonstrates some potency.[5] Thus, three of the five compounds in this series contain D-amino acids at position 5. The design rationale for compounds **4**, **26**, **24**, and **25** was explained previously. Given that both compounds **A** and **S** both exhibit cytotoxicity, compound **33**, which contains an *N*-methyl D-amino acid at position 5, provides the opportunity to explore the synergistic impact of a compound containing both structural features. Thus, our structural modifications at position 5 focused on the inclusion of D-amino acids and *N*-methyl moieties at position 5 in combination with structural elements at other positions.

Given that both D-amino acids and *N*-methyl moieties appear to play a key role in cytotoxicity,[4] we systematically explored the impact of these features on their biological activity in the first generation of compounds (Figure 9). Thus, the activities of San A peptide and **2**, its enantiomer, were compared, and both were found to have approximately the same antiproliferative activity.[5] Interestingly, both enantiomers in these enantiomeric pairs (**J** and **U**, **V** and **W**, **M** and **X**, **Y** and **Z**, and **AA** and **BB**) demonstrated the same activity. The only missing enantiomers were those paired to **A** and **CC**. Given that **A** was biologically active, the opposite enantiomer, **34**, was synthesized as part of this second generation of compounds.

Nine second-generation San A compounds were tested in the colon cancer cell line HT-29 (Figure 10), which is the same cancer cell line used for testing the first-generation compounds (Figure 2).[5] Comparison of San A peptide, which contains an L-phenylalanine in position 1, to compound **1**, which contains an L-tetrahydroisoquinoline residue, shows that the insertion of a rigid element in this position improves the percentage inhibition from 33 to 50%. Comparison of **G**, which has a D-phenylalanine at position 1, to compound **2**, which contains a D-tyrosine, shows that the addition of a hydrogen-bonding element has improved the percentage inhibition from 51 to 64%. These data suggest that perhaps the placement of a D-tetrahydroquinoline residue with a hydroxyl moiety at position 1 will increase the potency of that derivative.

For position 2, first-generation compounds **B** and **F** contain D-leucine in position 2, and both showed greater than 75% inhibition. Replacement of the D-leucine residue with a D-phenylalanine dramatically impacted the potency, dropping the percent inhibition from 75 to 35%. These data suggest that the binding pocket to which these San A derivatives bind cannot accommodate this more extended aromatic moiety. Derivatives with a smaller side chain may show greater potency than those with an isobutyl. Interestingly, first-generation compounds **F** and **E**, which contained *N*-methyl D-amino acids in positions 2 and 3, respectively, were both very potent (**E** = 77% and **F** = 89%), yet a combination of these two moieties, compound **8**, is not potent (48% inhibition). It appears that a specific conformational presentation is required for potency rather than having a particular moiety in a definitive position. This hypothesis seems reasonable given that recent studies show that a single *N*-methyl D-amino acid can potentially "lock" the conformation

of a macrocycle into a distinct structure, thus allowing it to bind to its biological target.[25,26]

We had observed that first-generation compounds containing a D-amino acid in position 3 tended to have reasonable potency (compounds **C** and **E**). Interestingly, compounds **N** and **O**, which contained methyl and ethyl moieties at this position, also exhibited reasonable potency, perhaps due to the smaller side chain in this position. To explore this concept at other positions, compound **17** was synthesized with a D-valine at position 3 (similar to **C**) but also an L-valine at positions 2, 4, and 5 rather than L-leucine. It appeared that this change was not favorable as the percent growth inhibition dropped from 82% (compound **C**) to 60% (compound **17**). In addition, an *N*-methyl D-moiety was placed in position 3 along with a cyclohexyl moiety (compound **21**). Comparison of the percentage inhibition between **E** and **21** indicates that the cyclohexyl moiety dramatically lowers the growth inhibition (from 89 to 22%, respectively). Finally, comparison of the percent inhibition of **C** to **26** (82 versus 41%, respectively) suggests that a D-valine in position 3 is not enough for potency, rather it also requires an alkyl moiety in position 5. These data support the idea that in order to achieve potency it is important for a specific conformational presentation of the side chains, and these data point to new structures that are useful for designing a third generation of compounds.

With regards to position 4, it appears that a large hydrophobic moiety at position 4 decreases the potency: **E** = 89% inhibition, and **21** = 22% inhibition. Compound **Q** (inhibition = 73%) with an *N*-methyl at position 4 and D-amino acid at position 5 was relatively potent. Yet comparison of **Q** to **32** (inhibition = 72%), which contains only an *N*-methyl at position 4, suggests that the D-amino acid at position 5 is not important for potency, and it is the *N*-methyl at position 4 that plays a significant role. Interestingly, **4**, which is almost identical to **Q** except it has a D-phenylalanine at position 1, has a much lower percent inhibition (15%) compared to that of **Q** (73%). These data indicate that a combination of selective D-amino acids, side chains, and *N*-methylation is not synergistic. Instead, these features play an individual role in the conformation of the macrocycle. Finally, it was determined that an *N*-methyl leucine in position 5 gave compound **A** with 77% inhibition, yet compound **26**, which contains an *N*-methyl glycine, only has 41% inhibition. Interestingly, compound **4**, which is a combination of **32** (72% inhibition), **G** (51% inhibition), and **S** (73% inhibition), gives a 15% inhibition! Thus, features shown to enhance potency in individual compounds are clearly not synergistic.

In summary, all compounds described in this paper are currently being tested on colon cancer cell line HT-29, as well as in six other cancer cell lines. These data are promising and strongly support the synthesis of a third generation of compounds. In addition, these initial SARs provide evidence that specific features play a key role in impacting the conformation of the macrocycles, which has a dramatic impact on their potency. Further studies on how conformation affects potency are underway and will be published in due course.

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Conclusion

San A derivatives have novel characteristics and are shown to target numerous cancers, including chemotherapeutically resistant colon and pancreatic cancers. As such, San A derivatives represent a new class of privileged structures with potent cytotoxic properties. Generation of the 34 structures described here explore the possible synergistic placement of D-amino acids in combination with four structural features (*N*-methyl amino acids, multiple aromatics, polar side chains, and alterations in the hydrophobic side chains). These compounds provide useful structure−activity relationships in drug-resistant cancers and highlight the key characteristics necessary for cytotoxicity. Furthermore, these second-generation compounds focus primarily on modifications demonstrated to increase potency in the first generation, which includes the addition of D-amino acids and *N*-methyl amino acids in positions 2, 3, and 5. This second generation also has an average C log P = 2.2 (versus 2.9 in the first generation),[15] and they incorporate four structural elements not previously introduced in any San A analogues.[27] These derivatives explore the role of stereochemistry in potency, examine the amide bond and transannular hydrogen-bonding influence using NH and *N*-methyl amino acids, and investigate the impact of polarity on cytotoxicity via substitution of aromatic, hydrophobic, and hydrophilic amino acids. The generation of these 34 second-generation compounds is a significant step forward in developing San A as a potential new class of drugs. Assays examining the cytotoxicity of these compounds are currently underway and will be reported in due course.

Experimental Methods

General Peptide Synthesis. All peptide coupling reactions were carried out under argon with dry solvent, using methylene chloride for dipeptide and tripeptide couplings and acetonitrile for pentapeptide couplings. The amine (1.1 equiv) and acid (1 equiv) were weighed into a dry flask along with 4 equiv of DIPEA and 1.1 equiv of TBTU. (Some coupling reactions would not go to completion using only TBTU, and therefore, ∼0.25 equiv of HATU and/or DEPBT was used. In a few cases, up to 1.1 equiv of all three coupling reagents was used.) Anhydrous methylene chloride was added to generate a 0.1 M solution. The solution was stirred at room temperature, and reactions were monitored by TLC. Reactions were run for 1 h before checking via TLC. If the reaction was not complete, an additional 0.25 equiv was of HATU and TBTU was added. If the reaction was complete, then workup was done by washing with saturated ammonium chloride. (Note that if acetonitrile was used for the reaction, methylene chloride was added to the reaction upon workup and the resulting solution was washed with ammonium chloride.) After back-extraction of aqueous layers with methylene chloride, organic layers were combined, dried over sodium sulfate, filtered, and concentrated. Flash chromatography using a gradient of ethyl acetate/hexane gave our desired peptide.

General Amine Deprotection. Amines were deprotected using 20% TFA in methylene chloride (0.1 M) with 2 equiv of anisole. The reactions were monitored by TLC, where the TLC sample was first worked up in a mini-workup using DI water and methylene chloride to remove TFA. Reactions were allowed to run for 1−2 h and then concentrated in vacuo.

General Acid Deprotection. Acids were deprotected using ∼4 equiv of lithium hydroxide (or until pH = ∼11) in methanol (0.1 M). The peptide was placed in a flask, along with lithium hydroxide

(27) See Supporting Information for all compound structures shown in the cytotoxicity assays.

and methanol and stirred overnight. Within 12 h, the acid was usually deprotected. Workup of reactions involved the acidification of the reaction solution using HCl to pH = 1. The aqueous solution was extracted three times with methylene chloride, and the combined organic layer was dried, filtered, and concentrated in vacuo.

Macrocyclization Procedure (in situ). All pentapeptides were acid- and amine-deprotected using concentrated HCl (8 drops per 0.3 mmol of linear pentapeptide) in THF (0.05 M). Anisole (2 equiv) was added to the reaction, and the reaction was stirred at room temperature. The reaction typically took 4 days, but TLC and LCMS were used to monitor the reaction every 12 h. LCMS data typically indicated the reaction was ~50% complete after the first day. Addition of 4 drops of concentrated HCl per 0.3 mmol of pentapeptide, stirring at rt overnight and checking the reaction via LCMS usually showed ~75% completion. On the fourth day, verification of the presence of the free amine and free acid and disappearance of the starting linear-protected pentapeptide permitted workup. The reaction was concentrated in vacuo, and the crude, dry, double deprotected peptide (free acid and free amine) was dissolved in a minimum solution of THF/acetonitrile/methylene chloride (2:2:1 ratio). Three coupling agents (DEPBT, HATU, and TBTU) were used at ~0.5−0.75 equiv each. These coupling agents were dissolved in a calculated volume of dry 40% THF, 40% acetonitrile, and 20% methylene chloride that would give a 0.004 M overall solution when included in the volume used for the deprotected peptide. The coupling agents were then added to the deprotected peptide solution. DIPEA (6 equiv or more in order to neutralize the pH) was then added to the reaction. The coupling agents are typically not very soluble in acetonitrile, which is why a combination of solvents is used.

After 1 h, TLC and LCMS (where the LCMS sample was worked up prior to injection) indicated that a product spot was developing. The comparison R_f value in the product spot on TLC was the protected linear pentapeptide. The reactions were always complete after 2 h, and monitoring the starting material deprotected pentapeptide via LCMS was the easiest method for determining completion. Upon completion, the reaction was worked up by washing with saturated ammonium chloride. After back-extraction of aqueous layers with large quantities of methylene chloride, the organic layers were combined, dried, filtered, and concentrated. All macrocycles were purified by initially running a crude plug of compound using an ethyl acetate/hexane gradient on silica gel, then running a column on the isolated product. Finally, when necessary, reverse phase HPLC was used for additional purification using a gradient of acetonitrile and DI water with 0.1% TFA.

Synthesis of Compound 1. Dipeptide 1c-2a. Dipeptide **1c-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 344 mg (1.8 mmol, 1.1 equiv) of amine **1c**, 408 mg (1.6 mmol, 1.0 equiv) of acid, 1.1 mL (4 equiv) of DIPEA, 395 mg (1.2 mmol, 0.75 equiv) of TBTU, and 467 mg (1.2 mmol, 0.75 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (635 mg, 92% yield): R_f 0.5 (EtOAc/Hex 3:4); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0−1.2 (m, 6H), 1.4 (s, 9H), 1.6 (m, 2H), 1.8−2.0 (m, 1H), 3.2−3.2 (m, 2H), 3.7 (s, 3H), 4.8−5.1 (s, 2H), 5.3 (br, αH), 5.5 (m, αH), 7.2−7.4 (m, 4H), 8.1 (m, 1H).

Dipeptide 1c-2a-NH$_2$. Dipeptide **1c-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (486 mg, 100% yield).

Tripeptide 1c-2a-3a. Tripeptide **1c-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 243 mg (0.75 mmol, 1.1 equiv) of amine **1c-2a**, 149 mg (0.68 mmol, 1.0 equiv) of acid, 0.6 mL (5 equiv) of DIPEA, and 275 mg (0.85 mmol, 1.25 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (253 mg, 71% yield): R_f 0.5 (EtOAc/Hex 3:4); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.1 (m, 12H), 1.5 (s, 9H), 1.6−1.8 (m, 1H), 2.1 (m, 2H), 3.2−3.4 (dd, 1H), 3.7 (s, 3H), 4.0−4.2 (br, 2H), 4.2 (m, αH), 5.0 (s, αH), 5.1 (s, αH), 5.2−5.4 (br, αH), 5.5 (m, 1H), 6.8 (d, 1H), 7.2−7.4 (m, 4H).

Tripeptide 1c-2a-3a-NH$_2$. Tripeptide **1c-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (204 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 401 mg (2.2 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.0 mmol, 1.0 equiv) of acid, 1.4 mL (8 equiv) of DIPEA, and 708 mg (2.2 mmol, 1.0 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (679 mg, 95% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.4 (s, 9H), 1.6−1.8 (t, 6H), 3.7 (s, 3H), 4.0−4.1 (dd, αH), 4.5−4.7 (m, αH), 4.8−4.9 (br, 1H), 6.4 (d, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (631 mg, 98% yield).

Pentapeptide 1c-2a-3a-4a-5a. Pentapeptide **1c-2a-3a-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 204 mg (0.48 mmol, 1.1 equiv) of amine **1c-2a-3a**, 114 mg (0.44 mmol, 1.0 equiv) of acid, 0.39 mL (5 equiv) of DIPEA, 106 mg (0.33 mmol, 0.75 equiv) of TBTU, and 125 mg (0.33 mmol, 0.75 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (120 mg, 37% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (400 MHz, CD_3OD) δ 0.7−0.9 (m, 24H), 1.4 (s, 9H), 1.5−1.7 (m, 6H), 3.0−3.1 (m, 2H), 3.5 (s, 3H), 4.1 (d, αH), 4.2 (d, αH), 4.4 (t, αH), 4.5−4.6 (m, 2αH), 4.6−4.7 (m, 2H), 5.0 (d, 1H), 7.1−7.3 (m, 4H), 7.4−7.5 (m, 1H), 7.5−7.6 (m, 1H), 7.6 (d, 1H).

Macrocycle 1c-2a-3a-4a-5a (Compound 1). Macrocycle **1c-2a-3a-4a-5a** (compound **1**) was synthesized following the Macrocyclization procedure, utilizing 92.7 mg (0.15 mmol, 1.0 equiv) of linear pentapeptide, 0.13 mL (5 equiv) of DIPEA, 48.5 mg (0.15 mmol, 1.0 equiv) of TBTU, 57.4 mg (0.15 mmol, 1.0 equiv) HATU, and 45.1 mg (0.15 mmol, 1.0 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (38 mg, 42% yield): R_f 0.5 (EtOAc/Hex 4:3); ^{1}H NMR (400 MHz, CD_3OD) δ 0.8−1.0 (m, 24H), 1.6−1.8 (m, 9H), 3.1−3.2 (m, 2H), 4.2 (m, αH), 4.3 (m, αH), 4.5 (t, αH), 4.7 (m, αH), 5.0 (m, αH), 5.2 (d, 1H), 7.2−7.3 (m, 4H), 7.3 (d, 1H), 7.5 (m, 1H), 7.7 (s, 1H); LCMS *m*/*z* calcd for $C_{33}H_{51}N_5O_5$ (M + 1) 598.39, found 598.3.

Synthesis of Compound 2. Dipeptide 1f-2a. Dipeptide **1f-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 614 mg (2.6 mmol, 1.1 equiv) of amine **1a**, 600 mg (2.4 mmol, 1.0 equiv) of acid, 1.7 mL (4 equiv) of DIPEA, and 865 mg (2.9 mmol, 1.1 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (880 mg, 89% yield): R_f 0.65 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (d, 6H), 1.4 (s, 9H), 1.6 (br, 1H), 2.0 (s, 2H), 3.0 (m, 2H), 3.7 (s, 3H), 4.1 (dd, αH), 4.8 (dd, αH), 6.7 (s, 1H), 6.8 (s, 1H), 6.8 (d, 2H), 7.0 (d, 2H), 7.2 (s, 1H).

Dipeptide 1f-2a-NH$_2$. Dipeptide **1f-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (664 mg, 100% yield).

Tripeptide 1f-2a-3a. Tripeptide **1f-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 664 mg (2.15 mmol, 1.1 equiv) of amine **1f-2a**, 425 mg (1.95 mmol, 1.0 equiv) of acid, 1.36 mL (4 equiv) of DIPEA, and 699 mg (2.3 mmol, 1.1 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (460 mg, 50**%** yield): R_f 0.65 (EtOAc/Hex 3:1); ^{1}H NMR (200 MHz, CD_3OD) δ 0.8−0.9 (m, 12H), 1.4 (s, 9H), 2.8−3.2 (m, 2H), 3.6 (s, 3H), 3.8 (d, αH), 4.4 (t, αH), 4.6 (dd, αH), 6.6−7.0 (dd, 4H).

Tripeptide 1f-2a-3a-NH$_2$. Tripeptide **1f-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This

dipeptide was taken on to the next reaction without further purification or characterization (355 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 400 mg (2.2 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.0 mmol, 1.0 equiv) of acid, 1.4 mL (4 equiv) of DIPEA, and 770.6 mg (2.4 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (660 mg, 97% yield); R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (dd, 12H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 3.7 (s, 3H), 4.0 (m, αH), 4.6 (m, αH), 4.8 (br, 1H), 6.4 (br, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (564 mg, 87.8% yield): ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.7−0.9 (dd, 12H), 1.2−1.6 (m, 2H), 1.4 (s, 9H), 2.0−2.2 (m, 2H), 3.2−3.3 (m, 2H), 3.6 (s, 3H), 3.8 (dd, αH), 4.2 (m, αH), 4.8 (quint, αH), 4.9 (br, 1H), 6.3 (br, 1H), 6.6 (br, 1H), 7.0−7.6 (m, 5H), 8.2 (br, 1H).

Pentapeptide 1f-2a-3a-4a-5a. Pentapeptide **1f-2a-3a-4a-5a** was synthesized following the General peptide Synthesis procedure, utilizing 355 mg (0.00087 mmol, 1.1 equiv) of amine **1f**, 416 mg (0.00079 mmol, 1.0 equiv) of acid, 0.55 mL (4 equiv) of DIPEA, and 283.4 mg (0.95 mmol, 1.2 equiv) of Depbt. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (252 mg, 43.4% yield): R_f 0.6 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8 (dd, 6H), 0.9−2.0 (m, 18H), 1.2−1.6 (m, 6H), 1.4 (s, 9H), 1.7 (m, 3H), 2.1 (m, 2H), 2.8−3.1 (m, 2H), 3.7 (s, 3H), 4.1 (m, 2αH), 4.4 (m, 2αH), 4.6 (m, αH), 6.7−7.0 (dd, 4H).

Macrocycle 1f-2a-3a-4a-5a (Compound 2). Macrocycle **1f-2a-3a-4a-5a** (compound **2**) was synthesized following the Macrocyclization procedure, utilizing 210.8 mg (0.34 mmol, 1.0 equiv) of linear pentapeptide, 0.48 mL (4 equiv) of DIPEA, 55.4 mg (0.17 mmol, 0.5 equiv) of TBTU, 68.2 mg (0.179 mmol, 0.5 equiv) HATU, and 51.4 mg (0.171 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase-HPLC to yield the macrocycle (8.0 mg, 5% yield): R_f 0.6 (EtOAc/Hex 1:0); ^{1}H NMR (500 MHz, CD_3OD) δ 0.6−0.8 (dd, 6H), 0.9−1.0 (m, 18H), 1.2−1.7 (m, 10H), 2.6−2.9 (m, 2H), 3.2 (m, αH), 4.1 (m, αH), 4.3 (m, αH), 4.4 (m, αH), 4.5 (m, αH), 6.7−7.0 (dd, 4H), 7.2 (d, 1H), 8.1 (d, 1H), 8.2 (d, 1H), 8.4 (d, 1H), 8.7 (d, 1H); LCMS *m*/*z* calcd for $C_{32}H_{51}N_5O_6$ (M + 1) 601.77, found 602.4.

Synthesis of Compound 3. Dipeptide 1e-2a. Dipeptide **1e-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 560.56 mg (2.2 mmol, 1.1 equiv) of amine **1e**, 500 mg (2.0 mmol, 1.0 equiv) of acid, 1.4 mL (4 equiv) of DIPEA, and 717.6 mg (2.39 mmol, 1.2 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (650 mg, 82% yield): R_f 0.65 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8 (d, 6H), 1.4 (s, 9H), 1.5−1.7 (m, 3H), 3.3 (d, 2H), 3.6 (s, 3H), 4.0 (m, αH), 4.9 (m, αH), 4.8 (br, 1H), 6.6 (br, 1H), 7.0−7.5 (m, 5H), 8.1 (br, 1H).

Dipeptide 1e-2a-NH$_2$. Dipeptide **1e-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (498 mg, 100% yield).

Tripeptide 1e-2a-3a. Tripeptide **1e-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 498 mg (1.5 mmol, 1.1 equiv) of amine **1e-2a**, 295.5 mg (1.36 mmol, 1.0 equiv) of acid, 0.95 mL (4 equiv) of DIPEA, and 488 mg (1.63 mmol, 1.2 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (488 mg, 67**%** yield): R_f 0.55 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.7−0.9 (dd, 12H), 1.2−1.6 (m, 2H), 1.4 (s, 9H), 2.0−2.2 (m, 2H), 3.2−3.3 (m, 2H), 3.6 (s, 3H), 3.8 (dd, αH), 4.2 (m, αH), 4.8 (quint, αH), 4.9 (br, 1H), 6.3 (br, 1H), 6.6 (br, 1H), 7.0−7.6 (m, 5H), 8.2 (br, 1H).

Tripeptide 1e-2a-3a-NH$_2$. Tripeptide **1e-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (399 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 799 mg (4.4 mmol, 1.1 equiv) of amine **4a**, 1 g (4.0 mmol, 1.0 equiv) of acid, 2.8 mL (4 equiv) of DIPEA, and 1.5 g (1.4 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (660 mg, 98% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (d, 12H), 1.4 (s, 9H), 1.5−1.8 (br, 6H), 3.7 (s, 3H), 4.1 (dd, αH), 4.6 (dd, αH), 4.9 (d, 1H), 6.4 (d, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (564 mg, 89% yield).

Pentapeptide 1e-2a-3a-4a-5a. Pentapeptide **1e-2a-3a-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 399 mg (0.92 mmol, 1.1 equiv) of amine **1e-2a-3a**, 371 mg (0.84 mmol, 1.0 equiv) of acid, 600 μL (4 equiv) of DIPEA, and 301 mg (1.01 mmol, 1.2 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (194 mg, 30% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 24H), 1.4 (m, 9H), 1.5−1.7 (m, 2H), 1.9−2.1 (d, 4H), 3.1 (m, 2H), 3.7 (s, 3H), 4.1 (d, 2αH), 4.3−4.4 (m, 2αH), 4.7 (m, αH), 7.0−7.1 (m, 4H), 7.2 (m, 1H), 7.3 (d, 1H), 7.5 (d, 1H), 7.8 (d, 1H), 8.0 (d, 1H).

Macrocycle 1e-2a-3a-4a-5a (Compound 3). Macrocycle **1e-2a-3a-4a-5a** (compound **3**) was synthesized following the Macrocyclization procedure, utilizing 167 mg (0.26 mmol, 1.0 equiv) of linear pentapeptide, 360 μL (8 equiv) of DIPEA, 42 mg (0.13 mmol, 0.5 equiv) of TBTU, 49.4 mg (0.13 mmol, 0.5 equiv) HATU, and 39 mg (0.13 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (5.5 mg, 33% yield): R_f 0.65 (EtOAc/MeOH 98:2); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 24H), 1.2−1.4 (m, 6H), 1.5−1.7 (m, 4H), 3.1−3.2 (m, 2H), 3.6−3.8 (m, 2αH), 4.0 (d, αH), 4.5 (m, αH), 4.6 (m, αH), 6.6 (d, 1H), 7.1−7.2 (m, 4H), 7.3 (d, 4H), 7.4 (m, 1H), 7.5 (d, 1H), 8.0 (d, 1H), 8.1 (d, 1H); LCMS *m*/*z* calcd for $C_{34}H_{52}N_6O_5$ (M + 1) 625.4, found 625.5.

Synthesis of Compound 4. Dipeptide 1b-2a. Dipeptide **1b-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 475 mg (2.2 mmol, 1.1 equiv) of amine **1b**, 500 mg (2.0 mmol, 1.0 equiv) of acid, 1.39 mL (4 equiv) of DIPEA, and 708 mg (2.2 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (721 mg, 92% yield): R_f 0.5 (EtOAc/Hex 35:65); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0 (d, 6H), 1.5 (s, 9H), 1.7 (m, 3H), 3.1−3.3 (sept, 2H), 3.8 (s, 3H), 4.2 (m, αH), 4.8 (br, 1H), 5.0 (q, αH), 6.6 (br, 1H), 7.2−7.4 (m, 5H).

Dipeptide 1b-2a-NH$_2$. Dipeptide **1b-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (537 mg, assume quantitative yield).

Tripeptide 1b-2a-3a. Tripeptide **1b-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 537 mg (1.83 mmol, 1.1 equiv) of amine **1b-2a**, 363 mg (1.67 mmol, 1.0 equiv) of acid **3a**, 1.17 mL (4 equiv) of DIPEA, and 590.4 mg (1.83 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (798 mg, 97% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.57 (s, 9H), 1.6−1.7 (m, 3H), 2.1−2.3 (sept, 1H), 3.1−3.4 (m, 2H), 3.8 (s, 3H), 3.9−4.0 (dd, αH), 4.5 (m, αH), 4.8−5.0 (q, αH), 5.0 (br, 1H), 6.4 (d, 1H), 6.6 (br, 1H), 7.2−7.4 (m, 5H).

Tripeptide 1b-2a-3a-NH$_2$. Tripeptide **1b-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This

dipeptide was taken on to the next reaction without further purification or characterization (635 mg, assume quantitative yield).

Dipeptide 4c-5b. Dipeptide **4c-5b** was synthesized following the General Peptide Synthesis procedure, utilizing 780 mg (3.99 mmol, 1.1 equiv) of amine **4c**, 904 mg (3.63 mmol, 1.0 equiv) of acid **5b**, 2.53 mL (4 equiv) of DIPEA, 875 mg (2.2 mmol, 0.75 equiv) of TBTU, and 690 mg (1.8 mmol, 0.5 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (140 mg, 15% yield): R_f 0.6 (EtOAc/Hex 35:65); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0−1.2 (m, 12H), 1.4−1.7 (m, 4H), 1.5 (s, 9H), 1.7−2.0 (m, 2H), 3.1 (s, 3H), 3.8 (s, 3H), 4.8 (br, αH), 5.3 (m, αH).

Dipeptide HO-4c-5b. Dipeptide **HO-4c-5b** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (133 mg, 99% yield).

Pentapeptide 1b-2a-3a-4c-5b. Pentapeptide **1b-2a-3a-4c-5b** was synthesized following the General Peptide Synthesis procedure, utilizing 192 mg (0.49 mmol, 1.1 equiv) of amine **1b-2a-3a**, 160 mg (0.44 mmol, 1.0 equiv) of acid **4c-5b**, 0.32 mL (4 equiv) of DIPEA, 78 mg (0.24 mmol, 0.55 equiv) of TBTU, and 93 mg (0.24 mmol, 0.55 equiv) HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (315 mg, 97% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.4 (s, 9H), 1.4−2.1 (m, 10H), 3.0 (s, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.4 (m, αH), 4.6 (m, αH), 4.8 (m, αH), 5.1 (m, αH), 7.1−7.3 (m, 5H).

Macrocycle 1b-2a-3a-4c-5b (Compound 4). Macrocycle **1b-2a-3a-4c-5b** (compound **4**) was synthesized following the Macrocyclization procedure, utilizing 200 mg (0.33 mmol, 1.0 equiv) of linear pentapeptide precursor, 0.35 mL (6 equiv) of DIPEA, 53 mg (0.16 mmol, 0.5 equiv) of TBTU, 63 mg (0.16 mmol, 0.5 equiv) of HATU, and 50 mg (0.16 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (7 mg, 3.6% yield): R_f 0.4 (EtOAc = 100%); LCMS *m*/*z* calcd for $C_{33}H_{53}N_5O_5$ (M + 1) 600.8, found 600.8.

Synthesis of Compound 6. Dipeptide 1a-2g. Dipeptide **1a-2g** was synthesized following the General Peptide Synthesis procedure, utilizing 690 mg (3.199 mmol, 1.2 equiv) of amine **1a**, 812 mg (2.908 mmol, 1.0 equiv) of acid **2g**, 2000 μL (4 equiv) of DIPEA, and 1120 mg (3.489 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1282 mg, 90% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.6 (s, 9H), 2.7 (s, 3H), 3.1 (br, 4H), 3.7 (s, 3H), 4.7−5.0 (br, 2αH), 6.6 (br, 1H) 7.0−7.4 (m, 10H).

Dipeptide 1a-2g-NH$_2$. Dipeptide **1a-2g-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (991 mg, quantitative yield).

Tripeptide 1a-2g-3a. Tripeptide **1a-2g-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 870 mg (2.557 mmol, 1.1 equiv) of amine **1a-2g-NH$_2$**, 505 mg (2.324 mmol, 1.0 equiv) of acid **3a**, 1600 μL (4 equiv) of DIPEA, and 895 mg (2.789 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (1250 mg, 72% yield): R_f 0.5 (EtOAc/Hex 13:7); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.5−0.7 (m, 6H), 1.4 (s, 9H), 1.6 (m, 1H), 2.9 (s, 3H), 3.1 (t, 4H), 3.6 (s, 3H), 4.2 (dd, αH), 4.8 (q, αH), 5.0 (br, 1H), 5.5 (dd, αH), 6.7 (br, 1H), 7.1−7.4 (m, 10H).

Tripeptide 1a-2g-3a-NH$_2$. Tripeptide **1a-2g-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1018 mg, quantitative yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 400 mg (2.206 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.006 mmol, 1.0 equiv) of acid **5a**, 1400 μL (4 equiv) of DIPEA, and 770 mg (2.399 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (720 mg, 99% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 4.8 (br, 1H), 6.5 (br, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (693 mg, 97% yield).

Pentapeptide 1a-2g-3a-4a-5a. Pentapeptide **1a-2g-3a-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 960 mg (2.173 mmol, 1.1 equiv) of amine **1a-2g-3a-NH$_2$**, 680 mg (1.975 mmol, 1.0 equiv) of acid **HO-4a-5a**, 2800 μL (8 equiv) of DIPEA, 440 mg (1.381 mmol, 0.7 equiv) of TBTU, and 520 mg (1.381 mmol, 0.7 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (900 mg, 60% yield): R_f 0.5 (EtOAc/Hex 3:2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.4−0.7 (dd, 6H), 0.9 (m, 12H), 1.2 (t, 3H), 1.4 (s, 9H), 1.6 (m, 4H), 2.9 (s, 3H), 3.0−3.2 (m, 4H), 3.7 (s, 3H), 4.0 (m, αH), 4.4−4.8 (m, 2αH), 4.9 (br, 1H), 5.6 (dd, αH), 6.4−6.6 (d, 2H), 7.0−7.4 (m, 10H).

Macrocycle 1a-2g-3a-4a-5a (Compound 6). Macrocycle **1a-2g-3a-4a-5a** (compound **6**) was synthesized following the Macrocyclization procedure, utilizing 169 mg (0.2590 mmol, 1.0 equiv) of linear pentapeptide, 450 μL (10 equiv) of DIPEA, 49 mg (0.1291 mmol, 0.5 equiv) of TBTU, 42 mg (0.1291 mmol, 0.5 equiv) of HATU, and 39 mg (0.1291 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (25 mg, 16% yield): R_f 0.5 (EtOAc/Hex 13:7); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.6−1.1 (m, 18H), 1.3−1.8 (m, 3H), 2.8−3.0 (m, 2H), 2.9 (s, 3H), 3.0−3.3 (m, 2H), 4.1 (br, αH), 4.5 (q, αH), 4.6 (br, αH), 5.2 (m, αH), 6.5−6.9 (br, 2H), 7.1−7.3 (m, 10H), 7.5 (br, 1H); LCMS *m*/*z* calcd for $C_{36}H_{51}N_5O_5$ (M + 1) 634.39, found 634.3.

Synthesis of Compound 7. Dipeptide 1a-2f. Dipeptide **1a-2f** was synthesized following the General Peptide Synthesis procedure, utilizing 630 mg (2.9 mmol, 1.1 equiv) of amine **1a**, 700 mg (2.6 mmol, 1.0 equiv) of acid, 1.8 mL (4 equiv) of DIPEA, and 1.0 g (3.2 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.1g, 98% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.4 (s, 9H), 2.0−3.1 (m, 4H), 3.6 (s, 3H), 4.3 (dd, αH), 4.7−4.9 (br, 1H), 4.8−4.9 (dd, αH), 6.3 (d, 1H) 6.9−7.3 (m, 10H).

Dipeptide 1a-2f-NH$_2$. Dipeptide **1a-2f-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (840 mg, 100% yield).

Tripeptide 1a-2f-3a. Tripeptide **1a-2f-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 840 mg (2.6 mmol, 1.1 equiv) of amine **1a-2f**, 510 mg (2.3 mmol, 1.0 equiv) of acid, 1.6 mL (4 equiv) of DIPEA, and 900 mg (2.8 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (1.2g, 91% yield): R_f 0.5 (EtOAc/Hex 5:3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.7−0.9 (dd, 6H), 1.4 (s, 9H), 2.0 (m, 1H), 2.9−3.1 (m, 4H), 3.6 (s, 3H), 3.9 (t, αH), 4.7−4.9 (m, 2H), 5.1 (d, αH), 6.5−6.6 (d, αH), 6.5−6.7 (br, 1H), 7.0−7.3 (m, 10H).

Tripeptide 1a-2f-3a-NH$_2$. Tripeptide **1a-2f-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (910 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 800 mg (4.4 mmol, 1.1 equiv) of amine **4a**, 1.0 g (4.0 mmol, 1.0 equiv) of acid, 2.8 mL (4 equiv) of DIPEA, and 1.5 g (4.8 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.4 g, 100% yield): R_f 0.5 (EtOAc/Hex 3:5); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0

(d, 12H), 1.4 (s, 9H), 1.5−1.8 (br, 6H), 3.7 (s, 3H), 4.1 (dd, αH), 4.6 (dd, αH), 4.9 (d, 1H), 6.4 (d, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1.3 g, 100% yield).

Pentapeptide 1a-2f-3a-4a-5a. Pentapeptide **1a-2f-3a-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 920 mg (2.2 mmol, 1.1 equiv) of amine **1a-2f-3a**, 680 mg (2.0 mmol, 1.0 equiv) of acid, 2.8 mL (8 equiv) of DIPEA, 440 mg (1.4 mmol, 0.7 equiv) of TBTU, and 520 mg (1.4 mmol, 0.7 equiv) HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (200 mg, 13% yield): R_f 0.5 (EtOAc/Hex 5:3); ^{1}H NMR (200 MHz, DMSO) δ 0.4−0.6 (dd, 6H), 0.8−1.0 (d, 12H), 1.4 (s, 9H), 1.5−1.8 (br, 4H), 2.9 (d, 1H), 3.6 (s, 3H), 3.9 (dd, αH), 4.1 (d, αH), 4.4 (d, αH), 4.5 (d, αH), 6.9 (d, 1H), 7.0−7.4 (m, 10H), 7.6 (d, 1H), 7.8 (d, 1H), 8.2 (d, 1H), 8.5 (d, 1H).

Macrocycle 1a-2f-3a-4a-5a (Compound 7). Macrocycle **1a-2f-3a-4a-5a** (compound **7**) was synthesized following the Macrocyclization procedure, utilizing 100 mg (0.16 mmol, 1.0 equiv) of linear pentapeptide, 0.28 mL (10 equiv) of DIPEA, 25 mg (0.08 mmol, 0.5 equiv) of TBTU, 35 mg (0.08 mmol, 0.5 equiv) HATU, and 23 mg (0.08 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (10 mg, 10% yield): R_f 0.5 (EtOAc/Hex 5:3); LCMS *m*/*z* calcd for $C_{35}H_{49}N_5O_5$ (M + 1) 620.37, found 620.3.

Synthesis of Compound 9. Dipeptide 1a-2e. Dipeptide **1a-2e** was synthesized following the General Peptide Synthesis procedure, utilizing 430 mg (2.03 mmol, 1.2 equiv) of amine **1a**, 500 mg (1.65 mmol, 1.0 equiv) of acid **2e**, 0.86 mL (3 equiv) of DIPEA, and 790 mg (2.47 mmol, 1.5 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (740 mg, 98% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.4 (s, 9H), 3.0−3.2 (sept, 2H), 3.4−3.8 (m, 2H), 3.6 (s, 3H), 4.2 (br, αH), 4.4 (dd, 2H), 4.8 (q, αH), 5.3 (br, 1H), 6.9 (br, 1H), 7.0−7.4 (m, 10H).

Dipeptide 1a-2e-NH$_2$. Dipeptide **1a-2e-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (578 mg, assume quantitative yield).

Tripeptide 1a-2e-3a. Tripeptide **1a-2e-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 1000 mg (2.8 mmol, 1.3 equiv) of amine **1a-2e**, 460 mg (2.1 mmol, 1.0 equiv) of acid, 1.47 mL (4 equiv) of DIPEA, 670 mg (2.1 mmol, 1.0 equiv) of TBTU, and 399 mg (1.05 mmol, 0.5 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (870 mg, 75% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (dd, 6H), 1.4 (s, 9H), 2.0−2.2 (sept, 1H), 3.0−3.2 (m, 2H), 3.4−4.0 (m, 2H), 3.6 (s, 3H), 3.8−4.0 (m, αH), 4.5 (dd, 2H), 4.6 (m, αH), 4.8 (q, αH), 5.0 (br, 1H), 6.7 (br, 1H), 6.9 (br, 1H), 7.0−7.4 (m, 10H).

Tripeptide 1a-2e-3a-NH$_2$. Tripeptide **1a-2e-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (713 mg, assume quantitative yield).

Dipeptide 4f-5d. Dipeptide **4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 920 mg (5.5 mmol, 1.2 equiv) of amine **4f**, 1000 mg (4.6 mmol, 1.0 equiv) of acid **5d**, 2.4 mL (3 equiv) of DIPEA, and 2210 mg (6.9 mmol, 1.5 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1440 mg, 94% yield): R_f 0.5 (EtOAc/Hex 35:65); ^{1}H NMR (200 MHz, $CDCl_3$) 0.9−1.0 (dd, 12H), 1.4 (s, 9H), 2.0−2.2 (sept, 2H), 3.7 (s, 3H), 3.8−4.0 (dd, αH), 4.5 (dd, αH), 5.0 (br, 1H), 6.4 (br, 1H).

Dipeptide HO-4d-5d. Dipeptide **HO-4f-5d** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1340 mg, 93% yield).

Pentapeptide 1a-2e-3a-4f-5d. Pentapeptide **1a-2e-3a-4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 400 mg (0.87 mmol, 1.1 equiv) of amine **1a-2e-3a**, 250 mg (0.79 mmol, 1.0 equiv) of acid **4f-5d**, 0.69 mL (5 equiv) of DIPEA, 152 mg (0.47 mmol, 0.6 equiv) of TBTU, and 300 mg (0.79 mmol, 1.0 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (310 mg, 52% yield): R_f 0.4 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.8 (dd, 18H), 1.4 (9H), 1.8−2.0 (m, 2H), 2.8−3.0 (m, 2H), 3.4 (d, 2H), 3.6 (s, 3H), 3.8 (t, αH), 4.2 (dd, αH), 4.3 (m, αH), 4.4 (s, 2H), 4.5 (m, αH), 4.6 (m, αH), 6.8 (d, 1H), 7.1−7.3 (m, 10H), 7.6 (d, 1H), 7.8 (d, 1H), 8.0 (d, 1H), 8.4 (d, 1H).

Macrocycle 1a-2e-3a-4f-5d (Compound 9). Macrocycle **1a-2e-3a-4f-5d** was synthesized following the Macrocyclization procedure, utilizing 150 mg (0.23 mmol, 1.0 equiv) of linear pentapeptide, 0.2 mL (5 equiv) of DIPEA, 73 mg (0.23 mmol, 1.0 equiv) of TBTU, and 87 mg (0.23 mmol, 1.0 equiv) of HATU. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (20 mg, 14% yield): R_f 0.5 (EtOAc/Hex 3:1); LCMS *m*/*z* calcd for $C_{34}H_{47}N_5O_6$ (M + 1) 622.77, found 621.9.

Synthesis of Compound 10. Dipeptide 1a-2e. Dipeptide **1a-2e** was synthesized following the General Peptide Synthesis procedure, utilizing 435 mg (1.19 mmol, 1.1 equiv) of amine **1a**, 500 mg (1.65 mmol, 1.0 equiv) of acid **2e**, 865 μL (4 equiv) of DIPEA, and 790 mg (2.48 mmol, 1.5 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (750 mg, 100% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.4 (s, 9H), 3.0−3.1 (m, 2H), 3.4−3.5 (q, 1H), 3.6 (s, 3H), 3.8 (m, 1H), 4.3 (br, αH), 4.4−4.5 (q, 2H), 4.8 (br, αH), 5.3 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 10H).

Dipeptide 1a-2e-NH$_2$. Dipeptide **1a-2e-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (586 mg, 100% yield).

Tripeptide 1a-2e-3a. Tripeptide **1a-2e-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 500 mg (1.40 mmol, 1.3 equiv) of amine **1a-2e-NH$_2$**, 230 mg (1.05 mmol, 1.0 equiv) of acid **3a**, 735 μL (4 equiv) of DIPEA, 335 mg (1.05 mmol, 1.0 equiv) of TBTU, and 200 mg (0.525 mmol, 0.5 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (467.3 mg, 80% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 6H), 1.4 (s, 9H), 2.0−2.2 (m, 1H), 3.0−3.2 (d, 2H), 3.5 (m, 1H), 3.7 (s, 3H), 3.9 (m, 2H), 4.4−4.5 (m, 2H), 4.6 (m, αH), 4.8 (m, αH), 5.0 (br, 1H), 6.7 (br, 1H), 6.9 (br, 1H), 7.0−7.3 (m, 10H).

Tripeptide 1a-2e-3a-NH$_2$. Tripeptide **1a-2e-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (383.3 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 400.5 mg (2.21 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.01 mmol, 1.0 equiv) of acid **5a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.21 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (721 mg, 100% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 4.8 (br, 1H), 6.4 (br, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (693.3 mg, 97% yield).

Pentapeptide 1a-2e-3a-4a-5a. Pentapeptide **1a-2e-3a-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 383.2 mg (1.05 mmol, 1.1 equiv) of amine **1a-2e-3a-NH$_2$**,

330 mg (1.05 mmol, 1.0 equiv) of acid **HO-4a-5a**, 492 μL (4 equiv) of DIPEA, 153 mg (0.475 mmol, 0.5 equiv) of TBTU, and 216.7 mg (0.570 mmol, 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (446.7 mg, 60% yield): R_f 0.7 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (br, 18H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 2.0−2.2 (m, 1H), 3.0−3.2 (m, 2H), 3.4−3.5 (m, 1H), 3.6 (s, 3H), 3.8 (m, 1H), 4.1−4.3 (br, 2αH), 4.4−4.5 (br, 2αH), 4.8 (br, αH), 5.1 (br, 1H), 5.3 (br, 1H), 6.7 (br, 1H), 6.9 (br, 1H), 7.1−7.4 (m, 10H).

Protected Macrocycle 1a-2e-3a-4a-5a. Macrocycle **1a-2e-3a-4a-5a** was synthesized following the Macrocyclization procedure, utilizing 200 mg (0.3 mmol, 1.0 equiv) of linear pentapeptide, 200 μL (4 equiv) of DIPEA, 48 mg (0.15 mmol, 0.5 equiv) of TBTU, 57 mg (0.15 mmol, 0.5 equiv) of HATU, and 45 mg (0.15 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (20 mg, 10% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 18H), 1.5−1.7 (m, 6H), 2.0 (m, 1H), 2.9 (m, 2H), 3.5 (m, 1H), 3.9 (m, αH), 4.1 (m, αH), 4.4 (m, αH), 4.5 (m, αH), 4.6 (m, αH), 7.1−7.4 (m, 10H), 7.7 (m, 1H), 7.9 (m, 1H), 8.3 (m, 1H), 8.6 (m, 1H), 8.7 (m, 1H); LCMS *m*/*z* calcd for $C_{36}H_{51}N_5O_6$ (M + 1) 650.37, found 650.60.

Deprotected Macrocycle 1a-2e-3a-4a-5a (Compound 10). Deprotected macrocycle **1a-2e-3a-4a-5a** (compound **10**) was synthesized utilizing 20 mg (0.03 mmol, 1.0 equiv) of protected macrocycle, 10% palladium−carbon (cat. amount). The crude reaction was purified by reverse phase HPLC to yield the deprotected macrocycle (20 mg, 10% yield; 8.0 mg, 95% yield): ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 18H), 1.6−1.8 (m, 6H), 2.0 (m, 1H), 2.9 (m, 2H), 3.6 (m, 1H), 3.9 (m, αH), 4.0 (m, αH), 4.2 (m, αH), 4.5 (m, αH), 5.5 (m, αH), 7.1−7.4 (m, 5H); LCMS *m*/*z* calcd for $C_{29}H_{45}N_5O_5$ (M + 1) 560.34, found 560.30.

Synthesis of Compound 11. Dipeptide 1a-2b. Dipeptide **1a-2b** was synthesized following the General Peptide Synthesis procedure, utilizing 475.8 mg (2.206 mmol, 1.1 equiv) of amine **1a**, 500 mg (2.006 mmol, 1.0 equiv) of acid **2b**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.206 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (654 mg, 83% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.5−4.7 (br, αH), 4.8−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2b-NH$_2$. Dipeptide **1a-2b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (486 mg, 100% yield).

Tripeptide 1a-2b-3e. Tripeptide **1a-2b-3e** was synthesized following the General Peptide Synthesis procedure, utilizing 486 mg (1.67 mmol, 1.1 equiv) of amine **1a-2b-NH$_2$**, 309 mg (1.52 mmol, 1.0 equiv) of acid **3e**, 1200 μL (4 equiv) of DIPEA, and 536.3 mg (1.67 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (590 mg, 81% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 9H), 1.4 (s, 9H), 1.5−1.7 (m, 4H), 1.8−2.0 (m, 1H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0 (m, αH), 4.4 (m, αH), 4.8 (m, αH), 4.9 (br, 1H), 6.4 (br, 1H), 6.6 (br, 1H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2b-3e-NH$_2$. Tripeptide **1a-2b-3e-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (458.3 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 401 mg (2.206 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.006 mmol, 1.0 equiv) of acid **5a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.206 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (620 mg, 90% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.6−1.8 (m, 6H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 4.8 (br, 1H), 6.4 (br, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (520 mg, 90% yield).

Pentapeptide 1a-2b-3e-4a-5a. Pentapeptide 1**a-2b-3e-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 457 mg (1.1 equiv) of amine **1a-2b-3e-NH$_2$**, 390 mg (1.13 mmol, 1.0 equiv) of acid **HO-4a-5a**, 800 μL (4 equiv) of DIPEA, 180 mg (0.56 mmol, 0.5 equiv) of TBTU, and 429.4 mg (1.13 mmol, 1.0 equiv) HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (557 mg, 64% yield): R_f 0.7 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 21H), 1.4 (s, 9H), 1.5−1.8 (m, 11H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.1 (m, 2αH), 4.4 (br, 2αH), 4.6 (m, αH), 7.1−7.3 (m, 5H).

Macrocycle 1a-2b-3e-4a-5a (Compound 11). Macrocycle **1a-2b-3e-4a-5a** (compound **11**) was synthesized following the Macrocyclization procedure, utilizing 233 mg (0.4 mmol, 1.0 equiv) of linear pentapeptide, 800 μL (4 equiv) of DIPEA, 64 mg (0.2 mmol, 0.5 equiv) of TBTU, 76 mg (0.2 mmol, 0.5 equiv) of HATU, and 60 mg (0.2 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (3 mg, 3% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 21H), 1.5−1.8 (m, 11H), 2.8 (m, 2H), 4.0 (m, αH), 4.1 (m, αH), 4.3 (m, αH), 4.5 (m, αH), 4.6 (m, αH), 7.2−7.4 (m, 5H), 7.6 (m, 1H), 7.8 (m, 1H), 8.0 (br, 1H), 8.2 (m, 1H), 8.6 (m, 1H); LCMS *m*/*z* calcd for $H_{31}H_{49}N_5O_5$ (M + 1) 572.37, found 572.3.

Synthesis of Compound 13. Macrocycle 1a-2e-3a-4f-5d (Compound 13). Deprotected macrocycle **1a-2e-3a-4f-5d** (compound **13**) was synthesized utilizing 10 mg (0.016 mmol, 1.0 equiv) of protected macrocycle, 10% palladium−carbon (cat. amount). The crude reaction was purified by reverse phase HPLC to yield the deprotected macrocycle (0.3 mg, 3.7% yield); LCMS *m*/*z* calcd for $C_{27}H_{41}N_5O_6$ (M + 1) 532.5, found 533.0.

Synthesis of Compound 16. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 475.8 mg (2.206 mmol, 1.1 equiv) of amine **1a**, 500 mg (500 mmol, 1.0 equiv) of acid **2a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.206 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (716.5 mg, 91% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.5−4.7 (br, αH), 4.8−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (534 mg, 100% yield).

Tripeptide 1a-2a-3f. Tripeptide **1a-2a-3f** was synthesized following the General Peptide Synthesis procedure, utilizing 500 mg (1.7 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 316 mg (1.55 mmol, 1.0 equiv) of acid **3f**, 1100 μL (4 equiv) of DIPEA, 550 mg (1.7 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (548 mg, 83% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 9H), 1.4 (s, 9H), 1.5−1.7 (m, 4H), 1.8−2.0 (m, 1H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0 (m, αH), 4.4 (m, αH), 4.8 (m, αH), 4.9 (br, 1H), 6.4 (br, 1H), 6.6 (br, 1H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2a-3f-NH$_2$. Tripeptide **1a-2a-3f-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (433.3 mg, 100% yield).

Dipeptide 4a-5a. Dipeptide **4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 401 mg (2.206

mmol, 1.1 equiv) of amine **4a**, 500 mg (2.01 mmol, 1.0 equiv) of acid **5a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.21 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (649.2 mg, 90% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.6−1.8 (m, 6H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 4.8 (br, 1H), 6.4 (br, 1H).

Dipeptide HO-4a-5a. Dipeptide **HO-4a-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (600 mg, 90% yield).

Pentapeptide 1a-2a-3f-4a-5a. Pentapeptide **1a-2a-3f-4a-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 376.7 mg (1.22 mmol, 1.1 equiv) of amine **1a-2a-3f-NH$_2$**, 345.6 mg (1.11 mmol, 1.0 equiv) of acid **HO-4a-5a**, 850 μL (4 equiv) of DIPEA, 177 mg (0.56 mmol, 0.5 equiv) of TBTU, and 421.8 mg (1.11 mmol, 1.0 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (466 mg, 60% yield): R_f 0.7 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 21H), 1.4 (s, 9H), 1.5−1.8 (m, 11H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.1 (m, 2αH), 4.4 (br, 2αH), 4.6 (m, αH), 7.1−7.3 (m, 5H).

Macrocycle 1a-2a-3f-4a-5a (Compound 16). Macrocycle **1a-2a-3f-4a-5a** (compound **16**) was synthesized following the Macrocyclization procedure, utilizing 191 mg (0.324 mmol, 1.0 equiv) of linear pentapeptide, 223 μL (4 equiv) of DIPEA, 42 mg (0.13 mmol, 0.4 equiv) of TBTU, 50 mg (0.13 mmol, 0.4 equiv) of HATU, and 40 mg (0.13 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (10 mg, 5.4% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 21H), 1.5−1.8 (m, 11H), 2.8 (m, 2H), 4.0 (m, αH), 4.1 (m, αH), 4.3 (m, αH), 4.5 (m, αH), 4.6 (m, αH), 7.2−7.4 (m, 5H), 7.6 (m, 1H), 7.8 (m, 1H), 8.0 (br, 1H), 8.2 (m, 1H), 8.6 (m, 1H); LCMS *m*/*z* calcd for $H_{31}H_{49}N_5O_5$ (M + 1) = 572.37, found 572.3.

Synthesis of Compound 17. Dipeptide 1a-2c. Dipeptide **1a-2c** was synthesized following the General Peptide Synthesis procedure, utilizing 1100 mg (5.06 mmol, 1.1 equiv) of amine **1a**, 1000 mg (4.6 mmol, 1.0 equiv) of acid **2c**, 3.2 mL (4 equiv) of DIPEA, and 1630 mg (5.06 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1700 mg, 98% yield): R_f 0.5 (EtOAc/Hex 35:65); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0 (dd, 6H), 1.5 (s, 9H), 2.2 (sept, 1H), 3.2 (dd, 2H), 3.8 (s, 3H), 4.0 (dd, αH), 5.0 (q, αH), 5.1 (br, 1H), 6.4 (br, 1H), 7.2−7.4 (m, 5H).

Dipeptide 1a-2c-NH$_2$. Dipeptide **1a-2c-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1250 mg, assume quantitative yield).

Tripeptide 1a-2c-3b. Tripeptide **1a-2c-3b** was synthesized following the General Peptide Synthesis procedure, utilizing 625 mg (2.24 mmol, 1.1 equiv) of amine **1a-2c**, 444 mg (2.04 mmol, 1.0 equiv) of acid **13b**, 1.4 mL (4 equiv) of DIPEA, and 720 mg (2.24 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (571 mg, 73% yield): R_f 0.4 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.6 (s, 9H), 2.2−2.4 (sept, 2H), 3.2 (sept, 2H), 3.8 (s, 3H), 4.0 (dd, αH), 4.4 (dd, αH), 5.0 (q, αH), 5.2 (br, 1H), 6.4−6.6 (br, 2H), 7.2−7.4 (m, 5H).

Tripeptide 1a-2c-3b-NH$_2$. Tripeptide **1a-2c-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (455 mg, assume quantitative yield).

Dipeptide 4f-5d. Dipeptide **4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 850 mg (5.06 mmol, 1.1 equiv) of amine **4f**, 1000 mg (6 mmol, 1.0 equiv) of acid **5d**, 3.21 mL (4 equiv) of DIPEA, and 1620 mg (5.06 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1400 mg, 92% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0−1.2 (dd, 12H), 1.6 (s, 9H), 2.2−2.4 (sept, 2H), 3.8 (s, 3H), 4.0 (dd, αH), 4.6 (dd, αH), 5.2 (br, 1H), 6.5 (br, 1H).

Dipeptide HO-4f-5d. Dipeptide **HO-4f-5d** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1300 mg, 97% yield).

Pentapeptide 1a-2c-3b-4f-5d. Pentapeptide **1a-2c-3b-4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 455 mg (1.15 mmol, 1.1 equiv) of amine **1a-2c-3b**, 330 mg (1.05 mmol, 1.0 equiv) of acid **4f-5d**, 0.75 mL (4 equiv) of DIPEA, 170 mg (0.52 mmol, 0.55 equiv) of TBTU, and 200 mg (0.52 mmol, 0.55 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (580 mg, 82% yield): R_f 0.8 (EtOAc/Hex 1:1); ^{1}H NMR (300 MHz, DMSO-d_6) δ 0.75−0.9 (m, 24H), 1.35 (s, 9H), 1.95 (br, 4H), 3.0 (sept, 2H), 3.55 (s, 3H), 3.7−3.8 (q, αH), 4.2 (t, 2αH), 4.3 (m, αH), 4.4 (q, αH), 7.1−7.3 (m, 5H).

Macrocycle 1a-2c-3b-4f-5d (Compound 17). Macrocycle **1a-2c-3b-4f-5d** (compound **17**) was synthesized following the Macrocyclization procedure, utilizing 79 mg (0.14 mmol, 1.0 equiv) of linear pentapeptide precursor, 0.15 mL (6 equiv) of DIPEA, 27 mg (0.08 mmol, 0.6 equiv) of TBTU, 32 mg (0.08 mmol, 0.6 equiv) of HATU, and 25 mg (0.08 mmol, 0.6 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (5 mg, 11% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.6−1.0 (m, 24H), 2.0 (m, 4H), 3.0−3.2 (m, 2H), 3.8 (m, αH), 4.1 (m, αH), 4.2 (m, αH), 4.5 (m, αH), 4.6 (m, αH), 7.2−7.4 (m, 5H); LCMS *m*/*z* calcd for $C_{29}H_{45}N_5O_5$ (M + 1) 544.34, found 542.2.

Synthesis of Compound 18. Dipeptide 1a-2c. Dipeptide **1a-2c** was synthesized following the General Peptide Synthesis procedure, utilizing 600 mg (2.25 mmol, 1.2 equiv) of amine **1a**, 500 mg (2.3 mmol, 1.0 equiv) of acid **2c**, 1.2 mL (3 equiv) of DIPEA, and 1100 mg (3.45 mmol, 1.5 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (770 mg, 88% yield): R_f 0.5 (EtOAc/Hex 35:65); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (dd, 6H), 1.4 (s, 9H), 2.0−2.2 (m, 1H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 3.8−4.0 (dd, αH), 4.8−4.9 (dd, αH), 4.9−5.0 (br, 1H), 6.2−6.3 (br, 1H).

Dipeptide 1a-2c-NH$_2$. Dipeptide **1a-2c-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (566 mg, assume quantitative yield).

Tripeptide 1a-2c-3h. Tripeptide **1a-2c-3h** was synthesized following the General Peptide Synthesis procedure, utilizing 590 mg (2.1 mmol, 1.3 equiv) of amine **1a-2c**, 480 mg (1.62 mmol, 1.0 equiv) of acid **3h**, 1.13 mL (4 equiv) of DIPEA, 480 mg (1.5 mmol, 0.8 equiv) of TBTU, and 300 mg (0.81 mmol, 0.8 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (730 mg, 81% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.7−0.9 (dd, 6H), 1.4 (s, 9H), 2.0−2.2 (m, 1H), 2.9−2.2 (m, 2H), 3.5−3.9 (m, 2H), 3.6 (s, 3H), 4.2−4.3 (dd, αH), 4.4−4.6 (dd, αH), 4.7−4.8 (dd, αH), 5.4 (br, 1H), 6.4 (br, 1H), 6.8 (br, 1H), 7.0−7.4 (m, 5H).

Tripeptide 1a-2c-3h-NH$_2$. Tripeptide **1a-2c-3h-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (598 mg, assume quantitative yield).

Dipeptide 4f-5d. Dipeptide **4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 920 mg (5.5 mmol, 1.2 equiv) of amine **4f**, 1000 mg (4.6 mmol, 1.0 equiv) of acid **5d**, 2.4 mL (3 equiv) of DIPEA, and 2210 mg (6.9 mmol, 1.5 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1440 mg, 94% yield): R_f 0.5 (EtOAc/Hex 35:65); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (dd, 6H), 1.2 (s, 9H), 2.1−2.3 (m, 2H), 3.7 (s, 3H), 3.8−4.0 (dd, αH), 4.5−4.6 (dd, αH), 5 (br, H), 6.4 (br, H).

Dipeptide HO-4f-5d. Dipeptide **HO-4f-5d** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1296 mg, 93% yield).

Pentapeptide 1a-2c-3h-4f-5d. Pentapeptide **1a-2c-3h-4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 500 mg (1.09 mmol, 1.1 equiv) of amine **1a-2c-3h**, 310 mg (1.0 mmol, 1.0 equiv) of acid **4f-5d**, 0.86 mL (5 equiv) of DIPEA, 190 mg (0.58 mmol, 0.6 equiv) of TBTU, and 370 mg (1.0 mmol, 1.0 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (670 mg, 45% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, DMSO-d_6) δ 0.7−0.8 (m, 18H), 1.4 (s, 9H), 1.8−2.0 (br, 3H), 2.9−3.0 (m, 2H), 3.5 (s, 3H), 3.8 (br, αH), 4.2 (br, αH), 4.3 (br, αH), 4.4 (br, αH), 4.4 (br, 2H), 4.7 (br, αH), 6.8 (br, 1H), 7.1−7.4 (m, 10H), 7.6 (br, 1H), 7.9 (br, 1H), 8.2 (br, 1H), 8.4 (br, 1H).

Macrocycle 1a-2c-3h-4f-5d (Compound 18). Macrocycle **1a-2c-3h-4f-5d** (compound **18**) was synthesized following the Macrocyclization procedure, utilizing 130 mg (0.2 mmol, 1.0 equiv) of linear pentapeptide precursor, 0.2 mL (6 equiv) of DIPEA, 80 mg (0.25 mmol, 1.2 equiv) of TBTU, 100 mg (0.26 mmol, 1.3 equiv) of HATU, and 80 mg (0.26 mmol, 1.3 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (10 mg, 8% yield): R_f 0.4 (EtOAc/Hex 1:1); LCMS *m*/*z* calcd for $C_{34}H_{47}N_5O_6$ (M + 1) 622.77, found 622.3.

Synthesis of Compound 19. Dipeptide 1a-2c. Dipeptide **1a-2c** was synthesized following the General Peptide Synthesis procedure, utilizing 1.1 g (5.06 mmol, 1.1 equiv) of amine **11a**, 1.0 g (4.60 mmol, 1.0 equiv) of acid, 3.2 mL (4 equiv) of DIPEA, and 1.63 g (5.06 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.7 g, 98% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 6H), 1.5 (s, 9H), 2.1−2.4 (m, 1H), 3.2 (d, 2H), 3.7 (s, 3H), 3.9−4.1 (m, αH), 4.9−5.1 (m, αH), 6.4−6.5 (br, 2H), 7.1−7.4 (m, 5H).

Dipeptide 1a-2c-NH$_2$. Dipeptide **1a-2c-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1.3 g, 100% yield).

Tripeptide 1a-2c-3f. Tripeptide **1a-2c-3f** was synthesized following the General Peptide Synthesis procedure, utilizing 626 mg (2.25 mmol, 1.1 equiv) of amine **1a-2c**, 415 mg (2.04 mmol, 1.0 equiv) of acid, 1.4 mL (4 equiv) of DIPEA, and 720 mg (2.24 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (900 mg, 95% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.1 (m, 9H), 1.5 (s, 9H), 1.7−2.0 (m, 2H), 3.0−3.2 (m, 1H), 3.1 (d, 2H), 3.7 (s, 3H), 3.8 (m, αH), 4.2 (m, αH), 4.8 (m, αH), 6.6 (br, 3H), 7.0−7.3−7.5 (m, 5H).

Tripeptide 1a-2c-3f-NH$_2$. Tripeptide **1a-2c-3f-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (706 mg, 100% yield).

Dipeptide 4f-5d. Dipeptide **4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 850 mg (5.01 mmol, 1.1 equiv) of amine **4f**, 1.0 g (4.60 mmol, 1.0 equiv) of acid, 3.2 mL (4 equiv) of DIPEA, and 1.63 g (5.06 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.4 g, 92% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0−1.2 (m, 12H), 1.5 (s, 9H), 1.8 (s, 1H), 2.1−2.4 (m, 1H), 3.9 (s, 3H), 4.0 (t, αH), 4.7 (t, αH), 5.2 (br, 1H), 6.5 (br, 1H).

Dipeptide HO-4f-5d. Dipeptide **HO-4f-5d** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1.3 g, 97% yield).

Pentapeptide 1a-2c-3f-4f-5d. Pentapeptide **1a-2c-3f-4f-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 353 mg (0.97 mmol, 1.1 equiv) of amine **1a-2c-3f**, 279 mg (0.88 mmol, 1.0 equiv) of acid, 0.8 mL (5 equiv) of DIPEA, 156 mg (0.49 mmol, 0.55 equiv) of TBTU, and 185 mg (0.49 mmol, 0.55 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (155 mg, 26% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, DMSO) δ 0.8−1.0 (m, 21H), 1.4 (s, 9H), 1.5−1.6 (m, 2H), 2.0 (m, 3H), 2.9−3.1 (m, 2H), 3.4 (s, 3H), 3.7−3.9 (m, αH), 4.0 (s, αH), 4.2 (m, αH), 4.3 (m, αH), 4.4 (m, αH), 6.9 (d, 1H), 7.1−7.3 (m, 5H), 7.6 (d, 1H), 7.8 (d, 1H), 8.1 (d, 1H), 8.4 (d, 1H).

Macrocycle 1a-2c-3f-4f-5d (Compound 19). Macrocycle **1a-2c-3f-4f-5d** (compound **19**) was synthesized following the Macrocyclization procedure, utilizing 184 mg (0.31 mmol, 1.0 equiv) of linear pentapeptide, 213 μL (4 equiv) of DIPEA, 48.2 mg (0.15 mmol, 0.5 equiv) of TBTU, and 174 mg (0.46 mmol, 1.5 equiv) of HATU. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (3 mg, 1.6% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.2 (m, 24H), 1.2−1.4 (m, 2H), 2.0−2.4 (m, 3H), 3.2 (m, 2H), 4.2 (m, αH), 4.8−5.0 (m, 2αH), 5.2 (m, αH), 5.3 (s, αH), 6.8 (d, 1H), 7.2−7.4 (m, 5H), 7.5 (m, 1H), 7.6 (m, 1H), 7.7 (m, 1H), 7.8 (m, 1H); LCMS *m*/*z* calcd for $C_{28}H_{43}N_5O_5$ (M − 1) 527.3, found 527.7.

Synthesis of Compound 20. Deprotected Macrocycle **1a-2c-3h-4f-5d** (compound **20**) was synthesized following the Hydrogenation procedure, utilizing 6 mg (0.009 mmol, 1.0 equiv) of protected macrocycle, 10% palladium−carbon (cat. amount). The crude reaction was purified by reverse phase HPLC to yield the deprotected macrocycle (0.31 mg, 6% yield); LCMS *m*/*z* calcd for $C_{27}H_{41}N_5O_6$ (M + 1) 532.5, found 532.5.

Synthesis of Compound 21. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 1904 mg (8.827 mmol, 1.1 equiv) of amine **1a**, 2000 mg (8.022 mmol, 1.0 equiv) of acid **2a**, 5,622 μL (4 equiv) of DIPEA, and 3090 mg (9.627 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (3115 mg, 99% yield): R_f 0.5 (EtOAc/Hex 7:13)' ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (2321 mg, quantitative yield).

Tripeptide 1a-2a-3d. Tripeptide **1a-2a-3d** was synthesized following the General Peptide Synthesis procedure, utilizing 631 mg (2.161 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 454 mg (1.962 mmol, 1.0 equiv) of acid **3d**, 1400 μL (4 equiv) of DIPEA, 378 mg (1.176 mmol, 0.6 equiv) of TBTU, and 448 mg (1.176 mmol 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (822 mg, 83% yield): R_f 0.5 (EtOAc/Hex 2:3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.5−1.8 (m, 2H), 2.2−2.4 (m, 1H), 2.7−2.9 (s, 3H), 3.1−3.2 (d, 2H), 3.7 (s, 3H), 4.0 (m, αH), 4.3 (m, αH), 4.8 (m, αH), 6.2 (br, 1H), 6.4−6.6 (br, 1H), 7.0−7.4 (m, 5H).

Tripeptide 1a-2a-3d-NH$_2$. Tripeptide **1a-2a-3d-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (649 mg, quantitative yield).

Dipeptide 4d-5a. Dipeptide **4d-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 488 mg (2.201 mmol, 1.1 equiv) of amine **4d**, 500 mg (2.006 mmol, 1.0 equiv) of acid **5a**, 1400 μL (4 equiv) of DIPEA, and 770 mg (2.399 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (729 mg, 92% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 6H), 1.1−1.4 (m, 5H), 1.5 (s, 9H), 1.6 (m,

6H), 1.7 (m, 4H), 1.9 (b, 1H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.5−4.7 (dd, αH), 4.8−4.9 (br, 1H), 6.3−6.4 (br, 1H).

Dipeptide HO-4d-5a. Dipeptide **HO-4d-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (637 mg, 94% yield).

Pentapeptide 1a-2a-3d-4d-5a. Pentapeptide **1a-2a-3d-4d-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 568 mg (1.401 mmol, 1.1 equiv) of amine **1a-2a-3d-NH$_2$**, 489 mg (1.273 mmol, 1.0 equiv) of acid, 1800 μL (8 equiv) of DIPEA, 339 mg (0.8901 mmol, 0.7 equiv) of HATU, and 190 mg (0.6402 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (833 mg, 83% yield): R_f 0.6 (EtOAc/Hex 7:3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 18H), 1.1−1.3 (m, 11H), 1.4 (s, 9H), 1.5−1.8 (m, 4H), 3.0 (s, 3H), 3.1 (d, 2H), 3.7 (s, 3H), 4.2−4.4 (dd, αH), 4.7−4.8 (dd, αH), 4.9−5.0 (br, 2αH), 5.8−6.0 (br, 1H), 6.4−6.5 (br, 1H), 6.7 (br, 1H), 7.1−7.3 (m, 5H).

Macrocycle 1a-2a-3d-4d-5a (Compound 21). Macrocycle **1a-2a-3d-4d-5a** (compound **21**) was synthesized following the Macrocyclization procedure, utilizing 302 mg (0.4610 mmol, 1.0 equiv) of linear pentapeptide, 680 μL (8 equiv) of DIPEA, 60 mg (0.1801 mmol, 0.4 equiv) of TBTU, 70 mg (0.1801 mmol, 0.4 equiv) of HATU, and 55 mg (0.1801 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (24 mg, 9% yield): R_f 0.65 (EtOAc/Hex 3:1); LCMS *m*/*z* calcd for $C_{36}H_{57}N_5O_5$ (M + 1) 640.44, found 640.4.

Synthesis of Compound 22. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 1904 mg (8.827 mmol, 1.1 equiv) of amine **1a**, 2000 mg (8.022 mmol, 1.0 equiv) of acid **2a**, 5622 μL (4 equiv) of DIPEA, and 3090 mg (9.627 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (3115 mg, 99% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (2321 mg, quantitative yield).

Tripeptide 1a-2a-3b. Tripeptide **1a-2a-3b** was synthesized following the General Peptide Synthesis procedure, utilizing 380 mg (1.301 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 257 mg (1.180 mmol, 1.0 equiv) of acid **3b**, 830 μL (4 equiv) of DIPEA, and 455 mg (1.421 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (539 mg, 92% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.57 (s, 9H), 1.6−1.7 (m, 3H), 2.1−2.3 (sept, 1H), 3.1−3.4 (m, 2H), 3.8 (s, 3H), 3.9−4.0 (dd, αH), 4.5 (m, αH), 4.8−5.0 (q, αH), 5.0 (br, 1H), 6.4 (d, 1H), 6.6 (br, 1H), 7.2−7.4 (m, 5H).

Tripeptide 1a-2a-3b-NH$_2$. Tripeptide **1a-2a-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (425 mg, quantitative yield).

Dipeptide 4e-5a. Dipeptide **4e-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 1460 mg (4.401 mmol, 1.1 equiv) of amine **4e**, 1000 mg (4.009 mmol, 1.0 equiv) of acid **5a**, 2800 μL (4 equiv) of DIPEA, 1030 mg (3.210 mmol, 0.8 equiv) of TBTU, and 610 mg (1.601 mmol, 0.4 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (2018 mg, 99% yield): R_f 0.5 (EtOAc/Hex 2:3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.2 (m, 6H), 1.6 (s, 9H), 1.7−2.2 (m, 10H), 3.3 (t, 1H), 3.5 (s, 3H), 4.3(m, αH), 4.6 (dd, αH), 5.3 (s, 2H), 7.4−7.6 (s, 5H).

Dipeptide HO-4e-5a. Dipeptide **HO-4e-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1921 mg, 98% yield).

Pentapeptide 1a-2a-3b-4e-5a. Pentapeptide **1a-2a-3b-4e-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 717 mg (1.831 mmol, 1.1 equiv) of amine **1a-2a-3b-NH$_2$**, 821 mg (1.661 mmol, 1.0 equiv) of acid **HO-4e-5a**, 2330 μL (8 equiv) of DIPEA, 267 mg (8.321 mmol, 0.5 equiv) of TBTU, 316 mg (8.321 mmol, 0.5 equiv) of HATU, and 100 mg (3.332 mmol, 0.2 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (705 mg, 49% yield): R_f 0.4 (EtOAc/Hex 3:2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 18H), 1.4 (s, 9H), 1.5−2.0 (m, 12H), 2.1−2.2 (m, 3H), 3.1−3.2 (m, 2H), 3.7 (s, 3H), 4.2−4.6 (br, 3αH), 4.8 (dd, αH), 5.1 (s, 2H), 5.3 (br, 1H), 6.2−6.6 (br, 3H), 6.9 (br, 3H), 7.1−7.4 (m, 10H).

Macrocycle 1a-2a-3b-4e-5a (Compound 22). Macrocycle **1a-2a-3b-4e-5a** (compound **22**) was synthesized following the Macrocyclization procedure, utilizing 613 mg (0.8130 mmol, 1.0 equiv) of linear pentapeptide, 1140 μL (8 equiv) of DIPEA, 104 mg (0.3250 mmol, 0.4 equiv) of TBTU, 124 mg (0.3250 mmol, 0.4 equiv) of HATU, and 97 mg (0.3250 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (10 mg, 2% yield): R_f 0.6 (MeOH/EtOAc 5:19); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.7−1.2 (m, 18H), 1.4−2.2 (m, 15H), 3.0−3.3 (m, 2H), 3.6−4.6 (m, 5αH), 5.0−5.1 (s, 2H), 6.8 (d, 1H), 7.1−7.4 (m, 10H); LCMS *m*/*z* calcd for $C_{40}H_{58}N_6O_7$ (M + 1) 735.44, found 736.5.

Synthesis of Compound 23. Macrocycle 1a-2a-3b-4e-5a (Compound 23). Macrocycle **1a-2a-3b-4e-5a** (compound **23**) was synthesized utilizing 10 mg (0.0136 mmol, 1.0 equiv) of macrocyclic pentapeptide, mg (cat. amount) of 10% palladium−carbon. The crude reaction was filtered using Celite-545 to yield the deprotected macrocycle (2 mg, 20% yield): ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.7−1.2 (m, 18H), 1.4−2.2 (m, 15H), 3.0−3.3 (m, 2H), 3.6−4.6 (m, 5αH), 5.1 (br, 1H), 5.5 (d, 1H), 6.8 (d, 1H), 7.1−7.4 (s, 5H), 7.6−7.8 (dd, 2H), 7.9−8.2 (m, 2H); LCMS *m*/*z* calcd for $C_{32}H_{52}N_6O_5$ (M + 1) 601.4, found 601.9.

Synthesis of Compound 24. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 1904 mg (8.827 mmol, 1.1 equiv) of amine **1a**, 2000 mg (8.022 mmol, 1.0 equiv) of acid **2a**, 5622 μL (4 equiv) of DIPEA, and 3090 mg (9.627 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (3115 mg, 99% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (2321 mg, quantitative yield).

Tripeptide 1a-2a-3b. Tripeptide **1a-2a-3b** was synthesized following the General Peptide Synthesis procedure, utilizing 631 mg (2.199 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 427 mg (1.899 mmol, 1.0 equiv) of acid **3b**, 1400 μL (4 equiv) of DIPEA, and 756 mg (2.399 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (800 mg, 86% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.57 (s, 9H), 1.6−1.7 (m, 3H), 2.1−2.3 (sept, 1H), 3.1−3.4 (m, 2H), 3.8 (s, 3H), 3.9−4.0 (dd, αH), 4.5 (m, αH), 4.8−5.0 (q, αH), 5.0 (br, 1H), 6.4 (d, 1H), 6.6 (br, 1H), 7.2−7.4 (m, 5H).

Tripeptide 1a-2a-3b-NH$_2$. Tripeptide **1a-2a-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (744 mg, quantitative yield).

Dipeptide 4a-5b. Dipeptide **4a-5b** was synthesized following the General Peptide Synthesis procedure, utilizing 802 mg (4.401

mmol, 1.1 equiv) of amine **4a**, 1000 mg (4.399 mmol, 1.0 equiv) of acid **5b**, 2800 μL (4 equiv) of DIPEA, and 1545 mg (4.799 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1210 mg, 85% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 4.8 (br, 1H), 6.5 (br, 1H).

Dipeptide HO-4a-5b. Dipeptide **HO-4a-5b** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1112 mg, 95% yield).

Pentapeptide 1a-2a-3b-4a-5b. Pentapeptide **1a-2a-3b-4a-5b** was synthesized following the General Peptide Synthesis procedure, utilizing 308 mg (0.9151 mmol, 1.1 equiv) of amine **1a-2a-3b-NH$_2$**, 286 mg (0.8310 mmol, 1.0 equiv) of acid **HO-4a-5b**, 600 μL (4 equiv) of DIPEA, 160 mg (0.4991 mmol, 0.6 equiv) of TBTU, and 190 mg (0.4991 mmol, 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (514 mg, 86% yield): R_f 0.6 (EtOAc/Hex 7:3); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.4 (s, 9H), 1.5−1.8 (m, 4H), 1.9−2.6 (m, 4H), 3.0−3.2 (dd, 2H), 3.7 (s, 3H), 4.2−5.0 (m, 5αH), 5.2 (br, 1H), 6.4−6.8 (br, 3H), 7.0−7.4 (m, 5H).

Macrocycle 1a-2a-3b-4a-5b (Compound 24). Macrocycle **1a-2a-3b-4a-5b** (compound **24**) was synthesized following the Macrocyclization procedure, utilizing 177 mg (0.2930 mmol, 1.0 equiv) of linear pentapeptide, 400 μL (8 equiv) of DIPEA, 38 mg (0.1210 mmol, 0.4 equiv) of TBTU, 45 mg (0.1210 mmol, 0.4 equiv) of HATU, and 35 mg (0.1210 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (86 mg, 51% yield): R_f 0.6 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.6−1.1 (m, 24H), 1.3−2.0 (m, 10H), 2.9−3.1 (m, 2H), 3.6 (m, αH), 3.9 (m, αH), 4.1−4.3 (m, 3αH), 7.1−7.3 (m, 5H); LCMS *m*/*z* calcd for $C_{32}H_{51}N_5O_5$ (M + 1) 586.39, found 586.2.

Synthesis of Compound 25. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 475.8 mg (2.21 mmol, 1.1 equiv) of amine **1a**, 500 mg (2.01 mmol, 1.0 equiv) of acid **2a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.21 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (779.5 mg, 99% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (580.9 mg, 100% yield).

Tripeptide 1a-2a-3b. Tripeptide **1a-2a-3b** was synthesized following the General Peptide Synthesis procedure, utilizing 580.9 mg (1.99 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 393.1 mg (1.91 mmol, 1.0 equiv) of acid **3b**, 1390 μL (4 equiv) of DIPEA, and 639 mg (1.99 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (890.6 mg, 95% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.4−1.6 (m, 3H), 2.0−2.2 (m, 1H), 3.1 (d, 2H), 3.7 (s, 3H), 3.9 (m, αH), 4.4 (m, αH), 4.8 (m, αH), 5.0 (br, 1H), 6.3 (br, 1H), 6.6 (br, 1H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2a-3b-NH$_2$. Tripeptide **1a-2a-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (709.2 mg, 100% yield).

Dipeptide 4a-5g. Dipeptide **4a-5g** was synthesized following the General Peptide Synthesis procedure, utilizing 570 mg (3.14 mmol, 1.1 equiv) of amine **4a**, 700 mg (2.85 mmol, 1.0 equiv) of acid **5g**, 2000 μL (4 equiv) of DIPEA, 550 mg (1.70 mmol, 0.6 equiv) of TBTU, and 650 mg (1.70 mmol, 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1058 mg, 99% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.5 (s, 9H), 1.6−1.8 (m, 6H), 2.7 (s, 3H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 6.2 (br, 1H), 6.4 (br, 1H).

Dipeptide HO-4a-5g. Dipeptide **HO-4a-5g** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1022 mg, 99% yield).

Pentapeptide 1a-2a-3b-4a-5g. Pentapeptide **1a-2a-3b-4a-5g** was synthesized following the General Peptide Synthesis procedure, utilizing 319 mg (0.81 mmol, 1.1 equiv) of amine **1a-2a-3b-NH$_2$**, 276 mg (0.74 mmol, 1.0 equiv) of acid **HO-4a-5g**, 520 μL (4 equiv) of DIPEA, 143 mg (0.44 mmol, 0.6 equiv) of TBTU, and 169 mg (0.44 mmol, 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (490 mg, 90% yield): R_f 0.6 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.4 (s, 9H), 1.5−1.7 (m, 9H), 1.9−2.0 (m, 1H), 2.3 (br, 1H), 2.8 (s, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.2 (br, αH), 4.3 (br, αH), 4.4 (m, 2αH), 4.8 (dd, αH), 6.4 (br, 1H), 6.6 (br, 1H), 6.7 (br, 1H), 6.9 (br, 1H), 7.1−7.3 (m, 5H).

Macrocycle 1a-2a-3b-4a-5g (Compound 25). Macrocycle **1a-2a-3b-4a-5g** (compound **25**) was synthesized following the Macrocyclization procedure, utilizing 353 mg (0.57 mmol, 1.0 equiv) of linear pentapeptide, 800 μL (4 equiv) of DIPEA, 173 mg (0.46 mmol, 0.8 equiv) of HATU, and 68 mg (0.23 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (31 mg, 10% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.5−1.7 (m, 9H), 2.1 (m, 1H), 2.6 (s, 3H), 3.0−3.2 (m, 2H), 3.9 (m, αH), 4.1 (m, αH), 4.4 (m, αH), 4.6 (dd, αH), 4.8 (m, αH) 5.1 (dd, 1H), 5.6−5.8 (dd, 1H) 6.2−6.4 (br, 1H), 7.0 (d, 1H), 7.1−7.3 (m, 5H); LCMS *m*/*z* calcd for $H_{33}H_{53}N_5O_5$ (M + 1) 600.40, found 600.60.

Synthesis of Compound 26. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 475.8 mg (2.21 mmol, 1.1 equiv) of amine **1a**, 500 mg (2.01 mmol, 1.0 equiv) of acid **2a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.21 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (779.5 mg, 99% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.6−1.8 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (br, αH), 4.7−4.8 (m, 1H), 4.8−4.9 (br, αH), 6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (580.9 mg, 100% yield).

Tripeptide 1a-2a-3b. Tripeptide **1a-2a-3b** was synthesized following the General Peptide Synthesis procedure, utilizing 580.9 mg (1.99 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 393.1 mg (1.91 mmol, 1.0 equiv) of acid **3b**, 1390 μL (4 equiv) of DIPEA, and 639 mg (1.99 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (890.6 mg, 95% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.5−1.7 (m, 3H), 2.0−2.2 (m, 1H), 3.1 (m, 2H), 3.7 (s, 3H), 3.9 (q, αH), 4.4 (m, αH), 4.8 (m, αH), 5.0 (br, 1H), 6.3 (br, 1H), 6.5 (br, 1H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2a-3b-NH$_2$. Tripeptide **1a-2a-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (709.2 mg, 100% yield).

Dipeptide 4a-5f. Dipeptide **4a-5f** was synthesized following the General Peptide Synthesis procedure, utilizing 528 mg (2.90 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.60 mmol, 1.0 equiv) of acid **5f**,

1850 μL (4 equiv) of DIPEA, and 1018 mg (3.20 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (775.1 mg, 100% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0 (s, 3H), 3.7 (s, 3H), 3.8−4.0 (q, 2αH) 4.6 (br, αH), 6.4 (br, 1H).

Dipeptide HO-4a-5f. Dipeptide **HO-4a-5f** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (738.7 mg, 100% yield).

Pentapeptide 1a-2a-3b-4a-5f. Pentapeptide **1a-2a-3b-4a-5f** was synthesized following the General Peptide Synthesis procedure, utilizing 560 mg (1.43 mmol, 1.1 equiv) of amine **1a-2a-3b-NH$_2$**, 369 mg (1.30 mmol, 1.0 equiv) of acid **HO-4a-5f**, 910 μL (4 equiv) of DIPEA, 250 mg (0.76 mmol, 0.6 equiv) of TBTU, and 297 mg (0.76 mmol, 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (660 mg, 88% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (200 MHz, CD_3OD) δ 0.9−1.1 (m, 18H), 1.5 (s, 9H), 1.6−1.7 (m, 6H), 2.1 (m, 1H), 3.0 (s, 3H), 3.1−3.2 (m, 2H), 3.7 (s, 3H), 3.8−4.0 (q, 2αH), 4.1 (m, αH), 4.4 (br, αH), 4.6 (br, αH), 4.7 (t, αH), 7.2−7.4 (m, 5H).

Macrocycle 1a-2a-3b-4a-5f (Compound 26). Macrocycle **1a-2a-3b-4a-5f** (compound **26**) was synthesized following the Macrocyclization procedure, utilizing 208 mg (0.37 mmol, 1.0 equiv) of linear pentapeptide, 260 μL (4 equiv) of DIPEA, 112 mg (0.30 mmol, 0.8 equiv) of HATU, and 44.3 mg (0.15 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (10 mg, 6.5% yield): R_f 0.4 (EtOAc/Hex 3:1); LCMS *m*/*z* calcd for $H_{29}H_{45}N_5O_5$ (M + 1) 544.34, found 545.30.

Synthesis of Compound 27. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 475.8 mg (2.206 mmol, 1.1 equiv) of amine **1a**, 500 mg (2.01 mmol, 1.0 equiv) of acid **2a**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.21 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (763.8 mg, 97% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.4 (s, 9H), 1.6−1.8 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (br, αH), 4.7−4.8 (br, αH), 4.8−4.9 (m, 1H), 6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (565.2 mg, 100% yield).

Tripeptide 1a-2a-3d. Tripeptide **1a-2a-3d** was synthesized following the General Peptide Synthesis procedure, utilizing 565.2 mg (1.95 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 408.2 mg (1.73 mmol, 1.0 equiv) of acid **3d**, 1300 μL (4 equiv) of DIPEA, and 626.1 mg (1.95 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (716 mg, 80% yield): R_f 0.6 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 2.2−2.4 (m, 1H), 2.8 (m, 3H), 3.1 (d, 2H), 3.7 (s, 3H), 4.0 (br, αH), 4.4 (br, αH), 4.8 (q, αH), 6.5−6.7 (br, 2H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2a-3d-NH$_2$. Tripeptide **1a-2a-3d-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (453.3 mg, 100% yield).

Dipeptide 4a-5b. Dipeptide **4a-5b** was synthesized following the General Peptide Synthesis procedure, utilizing 401 mg (2.206 mmol, 1.1 equiv) of amine **4a**, 500 mg (2.006 mmol, 1.0 equiv) of acid **5b**, 1400 μL (4 equiv) of DIPEA, and 708.3 mg (2.206 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (577.1 mg, 80% yield): R_f 0.7 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 3.7 (s, 3H), 4.1 (br, αH), 4.6 (br, αH), 4.8 (br, 1H), 6.5 (br, 1H).

Dipeptide HO-4a-5b. Dipeptide **HO-4a-5b** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (538 mg, 97% yield).

Pentapeptide 1a-2a-3d-4a-5b. Pentapeptide **1a-2a-3d-4a-5b** was synthesized following the General Peptide Synthesis procedure, utilizing 453.3 mg (1.12 mmol, 1.1 equiv) of amine **1a-2a-3d-NH$_2$**, 352 mg (1.08 mmol, 1.0 equiv) of acid **HO-4a-5b**, 754 μL (4 equiv) of DIPEA, 69.4 mg (0.22 mmol, 0.2 equiv) of TBTU, and 410.4 mg (1.08 mmol, 1.0 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (474.3 mg, 60% yield): R_f 0.7 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.5 (s, 9H), 1.5−1.7 (m, 9H), 2.3 (br, 1H), 2.9 (s, 3H), 3.1 (m, 2H), 3.7 (s, 3H), 4.1 (m, αH) 4.2 (dd, αH), 4.3 (br, αH), 4.5 (m, αH), 4.8 (dd, αH), 5.5 (br, 1H), 6.6 (br, 1H), 6.6 (br, 1H), 6.8 (br, 1H), 7.1−7.3 (m, 5H).

Macrocycle 1a-2a-3d-4a-5b (Compound 27). Macrocycle **1a-2a-3d-4a-5b** (Compound 27) was synthesized following the Macrocyclization procedure, utilizing 118 mg (0.191 mmol, 1.0 equiv) of linear pentapeptide, 134 μL (4 equiv) of DIPEA, 31 mg (0.096 mmol, 0.5 equiv) of TBTU, 36 mg (0.096 mmol, 0.5 equiv) of HATU, and 29 mg (0.096 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (10 mg, 8.42% yield): R_f 0.5 (EtOAc/Hex 3:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.4−1.7 (m, 9H), 2.2 (br, 1H), 3.0−3.2 (dd, 2H), 4.1 (dx, αH), 4.3 (br, αH), 4.4 (m, 2αH), 4.8 (dd, αH), 5.9 (d, 1H), 6.4 (br, 1H), 6.8 (br, 1H), 6.8 (br, 1H), 7.1−7.2 (m, 5H), 7.3 (br, 1H); LCMS *m*/*z* calcd for $H_{33}H_{53}N_5O_5$ (M + 1) 600.40, found 600.50.

Synthesis of Compound 28. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 1904 mg (8.827 mmol, 1.1 equiv) of amine **1a**, 2000 mg (8.022 mmol, 1.0 equiv) of acid **2a**, 5622 μL (4 equiv) of DIPEA, and 3090 mg (9.627 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (3115 mg, 99% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H)

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (2321 mg, quantitative yield).

Tripeptide 1a-2a-3a. Tripeptide **1a-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 360 mg (1.231 mmol, 1.1 equiv) of amine **1a-2a-NH$_2$**, 243 mg (1.121 mmol, 1.0 equiv) of acid **3a**, 800 μL (4 equiv) of DIPEA, and 431 mg (1.339 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (500 mg, 91% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.57 (s, 9H), 1.6−1.7 (m, 3H), 2.1−2.3 (sept, 1H), 3.1−3.4 (m, 2H), 3.8 (s, 3H), 3.9−4.0 (dd, αH), 4.5 (m, αH), 4.8−5.0 (q, αH), 5.0 (br, 1H), 6.4 (d, 1H), 6.6 (br, 1H), 7.2−7.4 (m, 5H).

Tripeptide 1a-2a-3a-NH$_2$. Tripeptide **1a-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (399 mg, quantitative yield).

Dipeptide 4e-5a. Dipeptide **4e-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 1460 mg (4.401 mmol, 1.1 equiv) of amine **4e**, 1000 mg (4.009 mmol, 1.0 equiv) of acid **5a**, 2,800 μL (4 equiv) of DIPEA, 1030 mg (3.210 mmol, 0.8 equiv) of TBTU, and 610 mg (1.601 mmol, 0.4 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (2018 mg, 99% yield): R_f 0.5 (EtOAc/Hex 2:3); ^{1}H NMR (200 MHz, $CDCl_3$) δ

1.2 (m, 6H), 1.6 (s, 9H), 1.7−2.2 (m, 10H), 3.3 (t, 1H), 3.5 (s, 3H), 4.3(m, αH), 4.6 (dd, αH), 5.3 (s, 2H), 7.4−7.6 (s, 5H).

Dipeptide HO-4e-5a. Dipeptide **HO-4e-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1921 mg, 98% yield).

Pentapeptide 1a-2a-3a-4e-5a. Pentapeptide **1a-2a-3a-4e-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 399 mg (1.021 mmol, 1.1 equiv) of amine **1a-2a-3a-NH$_2$**, 458 mg (0.9311 mmol, 1.0 equiv) of acid **HO-4e-5a**, 1300 μL (8 equiv) of DIPEA, 179 mg (0.5611 mmol, 0.6 equiv) of TBTU, and 212 mg (0.5611 mmol, 0.6 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (520 mg, 65% yield): R_f 0.4 (EtOAc/Hex 3:2); ^{1}H NMR (500 MHz, CD_3OD) δ 0.8−1.0 (m, 18H), 1.4 (s, 9H), 1.5−1.9 (m, 12H), 2.0−2.1 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 3.9−4.2 (dd, 3αH), 4.3−4.5 (br, 2H), 4.6 (dd, αH), 5.1 (s, 2H), 5.5 (d, 1H), 7.2−7.4 (m, 10H).

Macrocycle 1a-2a-3a-4e-5a (Compound 28). Macrocycle **1a-2a-3a-4e-5a** (compound **28**) was synthesized following the Macrocyclization procedure, utilizing 451 mg (0.5990 mmol, 1.0 equiv) of linear pentapeptide, 840 μL (8 equiv) of DIPEA, 228 mg (0.5990 mmol, 1.0 equiv) of HATU, and 36 mg (0.1190 mmol, 0.2 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (219 mg, 50% yield): R_f 0.6 (MeOH/EtOAc 5:19); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.6−1.2 (m, 18H), 1.3−2.3 (m, 15H), 3.0−3.3 (m, 2H), 3.5−4.6 (m, 5αH), 4.4.7−4.8 (br, 2H), 5.1 (s, 2H), 5.3−5.6 (br, 2H), 7.0−7.4 (m, 10H); LCMS *m*/*z* calcd for $C_{40}H_{58}N_6O_7$ (M + 1) 735.44, found 735.5.

Synthesis of Compound 29. Dipeptide 1b-2b. Dipeptide **1b-2b** was synthesized following the General Peptide Synthesis procedure, utilizing 925 mg (4.412 mmol, 1.1 equiv) of amine **1b**, 1000 mg (4.412 mmol, 1.0 equiv) of acid **2b**, 2780 μL (4 equiv) of DIPEA, and 1416 mg (4.412 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1572 mg, 99% yield): R_f 0.5 (EtOAc/Hex 7:13); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1b-2b-NH$_2$. Dipeptide **1b-2b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1172 mg, quantitative yield).

Tripeptide 1b-2b-3b. Tripeptide **1b-2b-3b** was synthesized following the General Peptide Synthesis procedure, utilizing 455 mg (1.556 mmol, 1.1 equiv) of amine **1b-2b-NH$_2$**, 307 mg (1.414 mmol, 1.0 equiv) of acid **3b**, 740 μL (3 equiv) of DIPEA, and 455 mg (1.556 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (640 mg, 92% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (m, 12H), 1.57 (s, 9H), 1.6−1.7 (m, 3H), 2.1−2.3 (sept, 1H), 3.1−3.4 (m, 2H), 3.8 (s, 3H), 3.9−4.0 (dd, αH), 4.5 (m, αH), 4.8−5.0 (q, αH), 5.0 (br, 1H), 6.4 (d, 1H), 6.6 (br, 1H), 7.2−7.4 (m, 5H).

Tripeptide 1b-2b-3b-NH$_2$. Tripeptide **1b-2b-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (510 mg, quantitative yield).

Dipeptide 4e-5a. Dipeptide **4e-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 1460 mg (4.401 mmol, 1.1 equiv) of amine **4e**, 1000 mg (4.009 mmol, 1.0 equiv) of acid **5a**, 2800 μL (4 equiv) of DIPEA, 1030 mg (3.210 mmol, 0.8 equiv) of TBTU, and 610 mg (1.601 mmol, 0.4 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (2018 mg, 99% yield): R_f 0.5 (EtOAc/Hex 2:3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.2 (m, 6H), 1.6 (s, 9H), 1.7−2.2 (m, 10H), 3.3 (t, 1H), 3.5 (s, 3H), 4.3(m, αH), 4.6 (dd, αH), 5.3 (s, 2H), 7.4−7.6 (s, 5H).

Dipeptide HO-4e-5a. Dipeptide **HO-4e-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1921 mg, 98% yield).

Pentapeptide 1b-2b-3b-4e-5a. Pentapeptide **1b-2b-3b-4e-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 383 mg (0.9710 mmol, 1.1 equiv) of amine **1b-2b-3b-NH$_2$**, 438 mg (0.8880 mmol, 1.0 equiv) of acid **HO-4e-5a**, 1250 μL (8 equiv) of DIPEA, 143 mg (0.4441 mmol, 0.5 equiv) of TBTU, 169 mg (0.4441 mmol, 0.5 equiv) of HATU, and 53 mg (0.1780 mmol, 0.2 equiv) of DEPBT. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (325 mg, 42% yield): R_f 0.4 (EtOAc/Hex 3:2); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 18H), 1.4 (s, 9H), 1.5−2.0 (m, 12H), 2.1−2.2 (m, 3H), 3.1−3.2 (m, 2H), 3.7 (s, 3H), 4.2−4.6 (br, 3αH), 4.8 (q, αH), 5.1 (s, 2H), 5.2−5.4 (br, 2H), 6.1 (br, 1H), 6.7 (br, 1H), 7.0−7.4 (m, 10H) 7.6 (br, 1H).

Macrocycle 1b-2b-3b-4e-5a (Compound 29). Macrocycle **1b-2b-3b-4e-5a** (compound **29**) was synthesized following the Macrocyclization procedure, utilizing 283 mg (0.3750 mmol, 1.0 equiv) of linear pentapeptide, 530 μL (8 equiv) of DIPEA, 48 mg (0.1510 mmol, 0.4 equiv) of TBTU, 57 mg (0.1510 mmol, 0.4 equiv) of HATU, and 45 mg (0.1510 mmol, 0.4 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (54 mg, 20% yield): R_f 0.6 (MeOH/EtOAc 5:19); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.7−1.2 (m, 18H), 1.4−2.2 (m, 15H), 3.0−3.3 (m, 2H), 3.6−4.6 (m, 5αH), 5.0−5.1 (s, 2H), 6.8 (d, 1H), 7.1−7.4 (m, 10H); LCMS *m*/*z* calcd for $C_{40}H_{58}N_6O_7$ (M + 1) 735.44, found 735.8.

Synthesis of Compound 30. Macrocycle 1a-2a-3a-4e-5a (Compound 30). Macrocycle **1a-2a-3a-4e-5a** (compound **30**) was synthesized utilizing 10 mg (0.0136 mmol, 1.0 equiv) of macrocyclic pentapeptide, mg (cat. amount) of 10% palladium−carbon. The crude reaction was filtered using Celite-545 to yield the deprotected macrocycle (8 mg, 80% yield): ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.7−1.2 (m, 18H), 1.4−2.2 (m, 15H), 3.0−3.3 (m, 2H), 3.6−4.6 (m, 5αH), 5.1 (br, 1H), 5.5 (d, 1H), 6.8 (d, 1H), 7.1−7.4 (s, 5H), 7.6−7.8 (dd, 2H), 7.9−8.2 (m, 2H); LCMS *m*/*z* calcd for $C_{32}H_{52}N_6O_5$ (M + 1) 601.4, found 601.1.

Synthesis of Compound 31. Macrocycle 1b-2b-3b-4e-5a (Compound 31). Macrocycle **1b-2b-3b-4e-5a** (compound **31**) was synthesized utilizing 54 mg (0.0717 mmol, 1.0 equiv) of macrocyclic pentapeptide, mg (cat. amount) of 10% palladium−carbon. The crude reaction was filtered using Celite-545 to yield the deprotected macrocycle (2 mg, 3.7% yield): LCMS *m*/*z* calcd for $C_{32}H_{52}N_6O_5$ (M + 1) 601.4, found 601.4.

Synthesis of Compound 32. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 670 mg (3.09 mmol, 1.1 equiv) of amine **1a**, 700 mg (2.81 mmol, 1.0 equiv) of acid, 2.0 mL (4 equiv) of DIPEA, and 1.08 g (3.37 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.07 g, 97% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (800 mg, 100% yield).

Tripeptide 1a-2a-3a. Tripeptide **1a-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 800 mg (2.73 mmol, 1.1 equiv) of amine **1a-2a**, 539 mg (2.48 mmol, 1.0 equiv) of acid, 2.3 mL (4 equiv) of DIPEA, 1.29 g (4.0 mmol, 1.2 equiv) of TBTU, and 0.5 equiv (0.5 mmol, 0.15 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (988 mg, 81% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.4−1.6 (m, 2H), 2.0−2.2 (m, 1H),

3.1 (d, 2H), 3.7 (s, 3H), 3.8 (m, αH), 4.2 (m, αH), 4.8 (m, αH), 5.0 (br, 1H), 6.2 (br, 1H), 6.3 (br, 1H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2a-3a-NH$_2$. Tripeptide **1a-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (787 mg, 100% yield).

Dipeptide 4c-5a. Dipeptide **4c-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 696 mg (3.55 mmol, 1.1 equiv) of amine **4c**, 805 mg (3.23 mmol, 1.0 equiv) of acid, 2.3 mL (4 equiv) of DIPEA, and 1.25 g (3.88 mmol, 1.2 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.02 g, 83% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 6H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 3.0 (s, 3H), 3.7 (s, 3H), 4.4 (br, αH), 4.7 (br, αH), 6.2 (br, 1H), 6.7 (br, 1H).

Dipeptide HO-4c-5a. Dipeptide **HO-4c-5a** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (892 mg, 89% yield).

Pentapeptide 1a-2a-3a-4c-5a. Pentapeptide **1a-2a-3a-4c-5a** was synthesized following the General Peptide Synthesis procedure, utilizing 787 mg (2.01 mmol, 1.1 equiv) of amine **1a-2a-3c**, 681 mg (1.82 mmol, 1.0 equiv) of acid, 3.5 mL (4 equiv) of DIPEA, 600 mg (1.74 mmol, 0.95 equiv) of TBTU, and 700 mg (1.74 mmol, 0.95 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (1.74 g, 96% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.4 (s, 9H), 1.6−1.8 (m, 9H), 2.2 (br, 1H), 2.8 (s, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.2 (m, αH), 4.4 (br, αH), 4.5 (b, αH), 4.8 (dd, αH), 5.0(m, 1H), 6.2 (br, 1H), 6.5 (br, 1H), 6.6 (br, 1H), 6.7 (br, 1H), 7.1−7.3 (m, 5H); LCMS *m*/*z* calcd for $C_{39}H_{65}N_5O_8$ (M + 23) 754.48, found 754.4.

Macrocycle 1a-2a-3a-4c-5a (Compound 32). Macrocycle **1a-2a-3a-4c-5a** (compound **32**) was synthesized following the Macrocyclization procedure, utilizing 153 mg (0.27 mmol, 1.0 equiv) of linear pentapeptide, 200 μL (10 equiv) of DIPEA, 40 mg (0.12 mmol, 0.5 equiv) of TBTU, 48 mg (1.24 mmol, 0.5 equiv) of HATU, and 40 mg (0.12 mmol, 0.5 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (22.3 mg, 14% yield): R_f 0.5 (EtOAc/Hex 1:1); LCMS *m*/*z* calcd for $C_{33}H_{53}N_5O_5$ (M + 1) 600.40, found 600.6.

Synthesis of Compound 33. Dipeptide 1a-2a. Dipeptide **1a-2a** was synthesized following the General Peptide Synthesis procedure, utilizing 476 mg (2.210 mmol, 1.1 equiv) of amine **1a**, 500 mg (2.010 mmol, 1.0 equiv) of acid, 1.4 mL (4 equiv) of DIPEA, and 774 mg (2.410 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (647 mg, 82% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.0 (dd, 6H), 1.5 (s, 9H), 1.5−1.7 (m, 3H), 3.0−3.2 (m, 2H), 3.7 (s, 3H), 4.0−4.2 (m, αH), 4.8−4.9 (m, αH), 4.7−4.9 (br, 1H), 6.4−6.5 (br, 1H) 7.1−7.4 (m, 5H).

Dipeptide 1a-2a-NH$_2$. Dipeptide **1a-2a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (483 mg, 100% yield).

Tripeptide 1a-2a-3a. Tripeptide **1a-2a-3a** was synthesized following the General Peptide Synthesis procedure, utilizing 483 mg (1.648 mmol, 1.1 equiv) of amine **1a-2a**, 325 mg (1.498 mmol, 1.0 equiv) of acid, 1.0 mL (4 equiv) of DIPEA, and 529 mg (1.648 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (568 mg, 77% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 12H), 1.4 (s, 9H), 1.4−1.6 (m, 2H), 2.0−2.2 (m, 1H), 3.1 (d, 2H), 3.7 (s, 3H), 3.8 (m, αH), 4.2 (m, αH), 4.8 (m, αH), 5.0 (br, 1H), 6.2 (br, 1H), 6.3 (br, 1H), 7.0−7.3 (m, 5H).

Tripeptide 1a-2a-3a-NH$_2$. Tripeptide **1a-2a-3a-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (451 mg, 100% yield).

Dipeptide 4a-5c. Dipeptide **4a-5c** was synthesized following the General Peptide Synthesis procedure, utilizing 602 mg (3.31 mmol, 1.1 equiv) of amine **4a**, 750 mg (3.01 mmol, 1.0 equiv) of acid, 2.1 mL (4 equiv) of DIPEA, and 116 mg (3.61 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (844 mg, 75% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.8−1.0 (m, 6H), 1.4 (s, 9H), 1.5−1.8 (m, 6H), 2.7 (s, 3H), 3.7 (s, 3H), 4.4 (br, αH), 4.7 (br, αH), 6.2 (br, 1H), 6.7 (br, 1H).

Dipeptide HO-4a-5c. Dipeptide **HO-4a-5c** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (509 mg, 63% yield).

Pentapeptide 1a-2a-3a-4a-5c. Pentapeptide **1a-2a-3a-4a-5c** was synthesized following the General Peptide Synthesis procedure, utilizing 451 mg (1.15 mmol, 1.1 equiv) of amine **1a-2a-3a**, 358 mg (1.05 mmol, 1.0 equiv) of acid, 1.5 mL (8 equiv) of DIPEA, 236 mg (0.74 mmol, 0.7 equiv) of TBTU, and 279 mg (0.74 mmol, 0.7 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (504 mg, 67% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.5−1.7 (s, 6H), 1.8 (m, 3H), 2.2 (br, 1H), 2.9 (s, 1H), 3.7 (s, 3H), 4.2 (m, αH), 4.3 (br, αH), 4.4 (br, αH), 4.6 (br, αH), 4.8 (m, αH), 6.2 (s, 1H), 6.6 (s, 1H), 6.7 (s, 1H), 6.9 (s, 1H), 7.1−7.4 (m, 5H); LCMS *m*/*z* calcd for $C_{33}H_{55}N_5O_6$ (M + 1) 618.82, found 618.4.

Macrocycle 1a-2a-3a-4a-5c (Compound 33). Macrocycle **1a-2a-3a-4a-5c** (compound **33**) was synthesized following the Macrocyclization procedure, utilizing 184 mg (0.31 mmol, 1.0 equiv) of linear pentapeptide, 213 μL (4 equiv) of DIPEA, 48.2 mg (0.15 mmol, 0.5 equiv) of TBTU, and 174 mg (0.46 mmol, 1.5 equiv) of HATU. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (3 mg, 1.6% yield): R_f 0.5 (EtOAc/Hex 1:1); LCMS *m*/*z* calcd for $C_{33}H_{53}N_5O_5$ (M + 1) 600.8, found 600.3.

Synthesis of Compound 34. Dipeptide 1b-2b. Dipeptide **1b-2b** was synthesized following the General Peptide Synthesis procedure, utilizing 952 mg (4.4 mmol, 1.1 equiv) of amine **1b**, 1.0 g (4.0 mmol, 1.0 equiv) of acid, 2.8 mL (4 equiv) of DIPEA, and 1.4 g (4.4 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the dipeptide (1.5 g, 98% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0−1.1 (dd, 6H), 1.5 (s, 9H), 1.8 (s, 3H), 3.2−3.4 (t, 2H), 3.8 (s, 3H), 4.1−4.3 (br, αH), 4.8−4.9 (m, αH), 4.8−5.0 (br, αH), 6.6 (d, 1H), 7.2−7.4 (m, 5H).

Dipeptide 1b-2b-NH$_2$. Dipeptide **1b-2b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (1.1 g, 100% yield).

Tripeptide 1b-2b-3b. Tripeptide 1b-2b-3b was synthesized following the General Peptide Synthesis procedure, utilizing 578 mg (2.0 mmol, 1.1 equiv) of amine **1b-2b**, 390 mg (1.8 mmol, 1.0 equiv) of acid, 1.25 mL (4 equiv) of DIPEA, and 634 mg (2.0 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the tripeptide (850 mg, 96% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.9−1.1 (d, 12H), 1.5 (s, 9H), 1.7−1.9 (m, 3H), 3.2 (d, 2H), 3.8 (s, 3H), 4.0 (br, αH), 4.5 (br, αH), 4.9 (dd, αH), 5.1 (d, 1H), 6.4 (d, 1H), 7.2−7.4 (m, 5H).

Tripeptide 1b-2b-3b-NH$_2$. Tripeptide **1b-2b-3b-NH$_2$** was synthesized following the General Amine Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (693 mg, 100% yield).

Dipeptide 4b-5d. Dipeptide **4b-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 407 mg (2.2 mmol, 1.1 equiv) of amine **4b**, 500 mg (2.0 mmol, 1.0 equiv) of acid, 1.4 mL (4 equiv) of DIPEA, and 720 mg (2.0 mmol, 1.1 equiv) of TBTU. The crude reaction was purified by column chromatog-

raphy (silica gel, EtOAc/Hex) to yield the dipeptide (760 mg, 99% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0−1.1 (m, 12H), 1.5 (s, 9H), 1.6−1.8 (m, 6H), 2.8 (s, 3H), 3.8 (s, 3H), 4.6−4.8 (br, αH).

Dipeptide HO-4b-5d. Dipeptide **HO-4b-5d** was synthesized following the General Acid Deprotection procedure. This dipeptide was taken on to the next reaction without further purification or characterization (700 mg, 97% yield).

Pentapeptide 1b-2b-3b-4b-5d. Pentapeptide **1b-2b-3b-4b-5d** was synthesized following the General Peptide Synthesis procedure, utilizing 346 mg (0.88 mmol, 1.1 equiv) of amine **1b-2b-3b**, 290 mg (0.80 mmol, 1.0 equiv) of acid, 0.7 mL (5 equiv) of DIPEA, 142 mg (0.44 mmol, 0.55 equiv) of TBTU, and 172 mg (0.44 mmol, 0.55 equiv) of HATU. The crude reaction was purified by column chromatography (silica gel, EtOAc/Hex) to yield the pentapeptide (500 mg, 85% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.8−1.0 (m, 24H), 1.5 (s, 9H), 1.6−1.7 (m, 9H), 2.2−2.4 (br, 1H), 2.7 (s, 3H), 3.1−3.2 (m, 2H), 3.2 (d, αH), 3.7 (s, 3H), 4.2 (m, αH), 4.5 (br, αH), 4.8 (m, αH), 6.6−6.7 (br, 1H), 6.8−7.0 (br, 1H), 7.1−7.4 (m, 5H).

Macrocycle 1b-2b-3b-4b-5d (Compound 34). Macrocycle **1b-2b-3b-4b-5d** (compound **34**) was synthesized following the Macrocyclization procedure, utilizing 202 mg (0.32 mmol, 1.0 equiv) of linear pentapeptide, 0.34 mL (6 equiv) of DIPEA, 58 mg (0.18 mmol, 0.55 equiv) of TBTU, 69 mg (0.18 mmol, 0.55 equiv) of HATU, and 54 mg (0.18 mmol, 0.55 equiv) of DEPBT. The crude reaction was purified by reverse phase HPLC to yield the macrocycle (40 mg, 21% yield): R_f 0.5 (EtOAc/Hex 1:1); ^{1}H NMR (500 MHz, CD_3OD) δ 0.9−1.1 (m, 24H), 1.5−1.7 (m, 9H), 2.0 (dd, 1H), 3.0 (m, 2H), 3.2 (m, αH), 4.0 (br, αH), 4.3 (m, αH), 4.5 (m, 1H), 4.6 (m, αH), 5.0 (t, 1H), 7.2−7.3 (m, 5H), 7.6 (m, 1H); LCMS *m*/*z* calcd for $C_{33}H_{53}N_5O_5$ (M + 1) 600.40, found 600.4.

Acknowledgment. We thank the NIH/MARC (5T34GM-08303) program for support of R.R. (2005/2006). We thank Pfizer, La Jolla, for equipment and financial donations as well as their fellowships to T.J.S. (SDSU−Pfizer summer 2005, and Pfizer SURF 2006). We thank the Howell Foundation for support for T.J.S. (Spring 2006) and E.P. (Spring 2005). We thank the NIH/NIGMS-MBRS-IMSD program for support of J.C. (Spring 2005−Spring 2006 NIH-5R25GM58906) and the U.S. Coast Guard for support of J.D.B. (2004−2006). We thank San Diego State University for financial support. S.R.M. was supported by NIH (AI058241-01).

Supporting Information Available: Data for compounds and their intermediates. This material is available free of charge via the Internet at http://pubs.acs.org.

JO061830J